Testing the Quality of Seeds for Farm and Garden

By R. H. Porter

AGRICULTURAL EXPERIMENT STATION
IOWA STATE COLLEGE OF AGRICULTURE
AND MECHANIC ARTS

BOTANY AND PLANT PATHOLOGY SECTION
FARM CROPS SUBSECTION
AGRONOMY SECTION

BUREAU OF PLANT INDUSTRY, SOILS AND AGRICULTURAL ENGINEERING
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Testing the Quality of Seeds for Farm and Garden[1]

By R. H. Porter[2]

The yield and quality of a crop to be harvested depend in part on the quality of the seed that is planted. Seed quality is a relative term. It is based on a number of conditions, the more important of which are purity of the seed as expressed in terms of freedom from weed seed and other impurities, viability, freedom from seed-borne organisms and ability to resist infection by soil-borne organisms. Many factors and conditions affect seed quality, including crop sequence, type and frequency of cultivation, climate, time of harvesting, genetic constitution of the seed, variety of seed planted, storage, age and treatment of seed. The purpose of this bulletin is to discuss some of the factors which affect seed quality, to outline methods employed in a seed laboratory to determine seed quality and to illustrate how the results of seed laboratory tests may be applied in the planting of seeds.

DEFINITIONS

It is necessary to define a few terms that appear frequently in this bulletin before taking up the several topics in detail.

PURITY OF SEED

Pure seed is a term commonly used by seed analysts, law enforcement officials, farmers and seedsmen. It generally implies freedom from such impurities as weed seeds, foreign or inert matter and crop seeds other than the kind being tested. Pure seed may also imply purity as to type or variety. Varietal purity is dependent on a system of seed certification involving the planting of pure lines or types supplemented by field, bin and laboratory inspection of the seed that is produced. Seed purity is expressed as percentage by weight and as such indicates the degree of freedom from impurities.

In seed laboratory practice it is necessary to consider the

[1] Projects 86 and 427 of the Iowa Agricultural Experiment Station.

[2] The author is indebted to the United States Department of Agriculture for the loan of illustrations in fig. 5-19. Insertion of the common name beneath the botanical name has been done for the convenience of the reader. Much of the data in tables 3 and 4 was also borrowed from the U. S. D. A. The illustrations in fig. 20 were made by Marie Corkle Lincoln and in fig. 21 and several others as indicated by George Morris. To both these artists special thanks are due for their assistance.

He is indebted to Prof. R. A. Fisher, also to Messrs. Oliver & Boyd, Ltd. of Edinburgh, for permission to reprint Distribution of Chi-square with Corresponding Probabilities and Degrees of Freedom from "Statistical Methods for Research Workers."

meaning of the term "seed" from both the botanical and commercial standpoints. Botanically, a seed is a ripened ovule which, in general, is one in which the egg nucleus has been fertilized with the nucleus from a pollen grain. In many plants this fertilization includes fusion of the endosperm nuclei with a second pollen nucleus, followed by a subdivision and growth of the resultant new cells until the embryo, the endosperm and the seed coats are fully developed. In nature some of the ovules of flowers of a given plant are never fertilized, or the young ovule fails to develop, yet the outer coverings are present and one cannot always differentiate such structures by external appearance from true seeds.

Commercially the term "seed" may include both seeds and fruits together with such accessory parts as the glumes of grasses, the old flower calyx and the involucre in the composite family. A fruit is a ripened ovary which may contain one or more ovules that become seeds. One-seeded fruits occur in such families as the grass, the sedge, the buckwheat and the sunflower (Compositae).

In the grass family the grain (fruit) is called a caryopsis in which the pericarp of the ovary is fused with the integuments (seed coats) of the ovule. The naked grains of wheat, corn, rye and hulless sorghum are typical caryopses or one-seeded fruits. Many grass "seeds" have additional coverings known as lemma or palea and in others such as unhulled sorghum and sudan grass, the covering consists of hard outer glumes with a papery lemma and palea inside and adjacent to the grain. In still other plants two or more fruits occur together as a bur (sand bur and buffalo grass).

In the buckwheat and sunflower families the one-seeded fruits are achenes in which the pericarp may be removed by hand or in seed cleaning processes. It is frequently impossible to determine, by external appearance, when these structures are viable or non-viable. Structures of sour dock, sheepsorrel, buckwheat, endive, chicory, dandelion and sunflower that appear to be fruits are often non-viable and have only an empty fruit case. Such structures are not seeds, they are inert material and it is unscientific to consider them as a part of pure seed or as weed seeds.

In the carrot family the fruit consists of two one-seeded carpels often attached together as in caraway, dill and celery. It is common for one member of the pair to be shriveled and undeveloped in which case it is not a seed or a fruit. In the carrot the members of each paired carpel usually separate at maturity, but some of them are infertile, they contain no embryo. They are neither true seeds nor fruits, yet they cannot be differentiated from true fruits by external appearance.

Possible disposition of these structures that are not true seeds or fruits will be considered in a later section.

SEED VIABILITY

Viability refers to the ability of seeds, when placed under favorable conditions, to germinate and produce plants. Viable seeds are those that possess a living embryo (germ) capable of initiating the processes of growth. Seed viability is determined by germination tests and is expressed as percentage by number of the pure seeds that are germinable.

SEED LONGEVITY

The length of life of seeds when stored in a laboratory or seed house as well as when mixed with field soil and exposed to natural conditions is expressed as "longevity." Seeds vary greatly in the number of years that viability may be retained.

SEED DORMANCY

Dormancy in seeds is a condition that prevents resumption of growth by a viable embryo when placed under conditions known to be favorable for germination of the kind of seed in question. It is caused by an unripe or undeveloped embryo or by an impermeable membrane surrounding the embryo which prevents the necessary exchange of gases. If properly treated, dormant seeds will germinate.

IMPERMEABLE SEEDS

Impermeability is a term applied to a seed whose coat prevents the entrance of water to the embryo. Such a seed cannot germinate until the seed coat is treated in some way to make it permeable to water. Impermeable seeds are common in the legume, mallow and morning-glory families and are generally referred to as "hard" seeds. Usually such seeds are viable. By many botanists impermeability is considered as one form of dormancy. In this bulletin it is considered separately from dormancy, because of common usage among seedsmen and farmers.

NOXIOUS WEED SEEDS

Most states and countries have seed laws for the control of seeds offered for sale. Seeds of weeds that are most destructive and harmful to agriculture are classed as noxious by legislative act. Sale of crop seed containing primary noxious weed seeds is usually prohibited, and the presence and rate of occurrence of secondary noxious weed seeds must be shown on the label.

SEED-BORNE ORGANISMS

Many crop diseases are caused by microscopic organisms, and frequently a specific causal agent is carried by the seed. Such organisms are referred to as "seed-borne," and often they can be detected by seed laboratory tests. Recommendations for control can then be made.

FACTORS AFFECTING SEED QUALITY

PURITY

The most important factors that determine the purity of seed offered for sale are (a) the purity as to kind or variety of the seed that is sown for the production of the crop to be harvested, (b) kinds of other viable crop or weed seeds in the soil in which the seed was grown, (c) type and frequency of cultivation given and (d) kind and efficiency of seed cleaning equipment that is available.

It is obvious that a crop of pure seed cannot be harvested unless pure seed of the kind is sown. For example, if one wishes to produce pure Tama oats for seed, the first step is to be certain that the seed planted is free from other varieties of oats and other kinds of crop seeds. Similarly, the presence of such weeds as wild buckwheat, wild rose, false strawberry and mustard in oats intended for seed usually prevents the production of a seed crop of oats free from such weed seeds. Seeds of Canada thistle, field bindweed and quackgrass often occur in seed lots of oats, barley and wheat and when such seed is sown it is not only difficult to produce a seed crop free from such weed seeds but the land may become so infested that eradication of the pests may be difficult and costly. The first step then in production of pure lots of seed is to sow pure seed.

Seeds of many weeds retain their vitality in the soil for many years. Seeds of sour dock, mustard, pigweed, smartweed, lamb's quarters, foxtail, butterprint and pennycress may live in the soil for 20 years or more as shown in table 1 (4, 10, 14, 16, 22). The longevity of weed seeds makes it difficult to always have access to clean land for the production of a seed crop, hence even though clean seed is sown the crop to be harvested may be infested. The kinds of weed seeds in a field determine the kind of a crop that may be sown with safety, if at all, for seed production. For example, it would be unwise to sow rape for seed production in a field infested with mustard or to sow flax in a field infested with either false flax or wild buckwheat. Flax is a poor competitor with weeds and should not be sown in fields where weeds have been allowed repeatedly to mature seed. Red clover and alfalfa for seed production should not be sown in a field known to be infested with dodder seed. Soybeans cannot com-

TABLE 1. LONGEVITY OF WEED SEEDS IN SOIL.

Family*	Botanical name of plant	Common name	Known yrs. of life	Authority
Gramineae (grass)	*Agropyron repens*	Quack grass	4	King
	Avena fatua	Wild oats	10 or more	Duvel & Goss
	Bromus secalinus	Cheat	Less than 5	Darlington
	Chaetochloa glauca	Yellow foxtail	20 or more	Duvel & Goss
	Chaetochloa verticillata	Bristly foxtail	20 or more	Duvel & Goss
	Chaetochloa virdis	Green foxtail	20 or more	Duvel & Goss
	Sporobolus cryptandrus	Dropseed	20 or more	Duvel & Goss
Cyperaceae (sedge)	*Cyperus esculentus*	Yellow nutgrass	20 or more	Duvel & Goss
Urticaceae (nettle)	*Cannabis sativa*	Wild hemp	0	Duvel & Goss
Polygonaceae (buckwheat)	*Polygonum hydropiper*	Water pepper	50 or more	Darlington
	Polygonum persicaria	Smartweed	20 or more	Duvel & Goss
	Rumex crispus	Sour dock	40 or more	Darlington
	Rumex obtusifolius	Bitter dock	20 or more	Duvel & Goss
Chenopodiaceae (goosefoot)	*Chenopodium album*	Lambs quarter	40	Darlington
	Chenopodium hybridum	Maple-leaved goosefoot	20 or more	Duvel & Goss
Amaranthaceae (pigweed)	*Amaranthus retroflexus*	Pigweed	40 or more	Darlington
	Amaranthus graecizans	Tumbling pigweed	40 or more	Darlington
Coryophyllaceae (cockle)	*Agrostemma gilhago*	Corn cockle	0	Duvel & Goss
	Alsine media	Chickweed	30	Darlington
Portulacaceae (purslane)	*Portulacca oleracea*	Purslane	50 or more	Darlington
Nymphaeaceae (water lily)	*Nelumbo nuciferat*	Lotus	120 or more	Ogha
Cruciferae (mustards)	*Brassica nigra*	Black mustard	50 or more	Darlington
	Capsella bursa-pastoris	Shepherd's purse	35	Darlington
	Erysimum cheiranthoides	Wormseed mustard	15	Duvel & Goss
	Hymenophysa pubescens	White top	3 or more	Brown & Porter
	Lepidium draba	Pepper cress	2	Brown & Porter
	Lepidium repens	Hoary cress	3 or more	Brown & Porter
	Lepidium virginicum	Pepper grass	40 or more	Darlington
	Sisymbrium altissimum	Tumbling mustard	10	Duvel & Goss
	Thlaspi arvense	Pennycress	20 or more	Duvel & Goss
Rosaceae (rose)	*Potentilla monspeliensis*	Cinquefoil	20 or more	Duvel & Goss
Malvaceae (mallow)	*Abutilon abutilon*	Button weed	20 or more	Duvel & Goss
	Malva rotundifolia	Mallow	20 or more	Duvel & Goss
Euphorbiaceae (spurge)	*Chamaesyce maculata*	Prostrate spurge	Less than 5	Darlington
	Euphorbia esula	Leafy spurge	4 or more	Brown & Porter
Onagraceae (evening primrose)	*Oenothera biennis*	Evening primrose	50 or more	Darlington
Convolvulaceae (morning glory)	*Convolvulus arvensis*	Field bindweed	4 or more	Brown & Porter
	Convolvulus sepium	Hedge bindweed	20 or more	Duvel & Goss
	Cuscuta epilinum	Flax dodder	10	Duvel & Goss
	Cuscuta polygonorum	Dodder	20 or more	Duvel & Goss
Verbenaceae (vervain)	*Verbena hastata*	Blue vervain	20 or more	Duvel & Goss
	Verbena urticifolia	White vervain	20 or more	Duvel & Goss
Solanaceae (nightshade)	*Datura tatula*	Jimson weed	20 or more	Duvel & Goss
	Solanum carolinense	Horse nettle	11 or more	King
	Solanum elaegnifolium	White horse nettle	3 or more	Duvel & Goss
	Solanum nigrum	Black nightshade	20 or more	Duvel & Goss
Scrophulariaceae (figwort)	*Verbascum blattaria*	Mullein	50 or more	Darlington
Plantaginaceae (plantain)	*Plantago lanceolata*	Buckhorn	10	Duvel & Goss
	Plantago major	Common plantain	20 or more	Duvel & Goss
	Plantago rugellii	Rugel's plantain	20 or more	Duvel & Goss
Compositae (sunflower, aster and ragweed)	*Ambrosia artimisiaefolia*	Small ragweed	20 or more	Duvel & Goss
	Ambrosia trifida	Giant ragweed	20 or more	Duvel & Goss
	Anthemis cotula	Dog fennel	25	Duvel & Goss
	Arctium lappa	Burdock	20 or more	Duvel & Goss
	Carduus arvensis	Canada thistle	20 or more	Duvel & Goss
	Centaurea calcitrapa	Purple star thistle	3 or more	Brown & Porter
	Centaurea repens	Russian knapweed	3 or more	Brown & Porter
	Centaurea solstitialis	Barnaby's thistle	3 or more	Brown & Porter
	Chrysanthemum leucanthemum	Oxeye daisy	20 or more	Duvel & Goss
	Grindelia squarrosa	Gum weed	10	Duvel & Goss
	Helsanthus annuus	Wild sunflower	1	Duvel & Goss
	Lactuca scariola	Wild lettuce	3	Duvel & Goss
	Rudbeckia hirta	Black-eyed Susan	20 or more	Duvel & Goss
	Xanthium pennsylvanicum	Cockle bur	15	Duvel & Goss

* Arranged by families according to Gray.
† Not a weed. The oriental lotus plant.

pete successfully with butterprint, cockle bur or horse nettle and since the fruits of cockle bur and horse nettle are difficult to remove from soybean seed it is unwise to plant beans in a field known to be infested with such weeds. Oats and barley cannot compete with field bindweed which is one of our most destructive weeds. Futhermore, it is almost impossible to remove bindweed seeds from small grain seed by cleaning machinery.

Fields intended for production of seed should be handled one or more years in advance of seeding in such a way as to reduce the weed population to a minimum. This may be accomplished by (a) frequent harrowing of the soil prior to planting so as to encourage weed seed germination followed by destruction of seedlings and (b) cutting, pulling or covering weeds before the seed has ripened. Row crops such as corn, soybeans, garden beans and peas and many vegetable crops may be kept free of weeds between the rows by machine cultivation, but hand weeding within the rows is necessary.

The development of seed cleaning machinery within the past 25 years has been remarkable. It is now possible to remove from clover, alfalfa, timothy, flax and many other kinds of crop seeds most kinds of weed seeds which a quarter century ago would have made the same seed unsalable on the basis of present laws. All large capacity seed houses that buy home-grown seed direct from the farm have an extensive array of machinery unknown until recent years. Buckhorn seeds can now be removed from clover seed, dodder can be removed from clover, alfalfa, flax and lespedeza seed and even horse nettle seeds can be removed from lespedeza seed. A machine that removes catchfly and cockle seeds from alsike clover has been developed. Clover, alfalfa and timothy seed can be cleaned to a purity of at least 99 to 99.5 percent. The operation of such machinery is expensive and there is considerable loss in bulk weight of seed, hence dockage is necessary. Farmers are not equipped with the necessary machinery for cleaning most crops and must rely on seed houses or specially equipped centers.

Seeds produced by farmers may be either sold to a processor on the basis of a dockage which depends on the amount and kind of impurities to be removed, or consigned for cleaning on the basis of a given charge per 100 pounds of the uncleaned bulk lot.

VIABILITY

Factors that determine the viability of seeds are (a) stage of maturity at the time of harvest, (b) conditions to which seeds are exposed while developing, (c) conditions under which seeds are stored, (d) age of the seed, (e) heredity, (f) fungi carried by seeds, and (g) insects and nematodes of stored seeds.

Mature, well ripened seed possesses reserve food materials

and usually has greater vitality than immature seed. Harvesting seed when it is immature is generally a poor practice because it often results in shrivelled and partially developed seeds. Furthermore, the ability of immature or partially developed embryos to withstand adverse field conditions is less than for those that are well ripened and possess reserve food materials.

Seeds may be mature physiologically before the moisture content has been reduced to a point where they can be safely stored. In such cases artifical drying may be practiced, as is done in the processing of hybrid seed corn.

Inasmuch as facilities for artificial drying are limited and the practice is expensive, most seed crops must mature on the plants and be harvested when the moisture content is low enough to insure against heating and spoilage.

Climatic conditions and methods of harvesting, while the crop is being grown or harvested, often seriously affect the viability of seeds. One of the most important environmental factors is temperature. The vitality of corn is frequently lowered by early frosts when the moisture content of the kernels is high. Studies made by the Nebraska Agricultural Experiment Station (21) have shown that open-pollinated corn with 40 to 45 percent moisture was seriously lowered in vitality by a temperature of 28°F. At temperatures of 24 to 20°F. and 16 to 12°F. the vitality of seed corn was lowered if the moisture content of the seed was below 25 and 20 percent, respectively.

Tests at the Iowa Station with seed of hybrid corn were made in 1942, using samples collected after the frost of Sept. 27, when temperatures ranged between 18 and 22°F. Only two lots, one with 40, and the other with 52 percent moisture, were injured. Several lots with 30 to 50 percent moisture did not give maximum germination at the time of collection, but when the moisture content had been reduced to 25 percent or less, germination of 90 to 95 percent occurred readily. These results indicate that some lots of hybrid seed corn with a given moisture content may be able to withstand lower temperatures than open-pollinated corn and that the extent of injury from exposure to low temperatures cannot be determined immediately.

Seed of eight varieties of soybean and nine varieties of sorghum was harvested in the fall of 1941 (43) at the Iowa Station at intervals, and the moisture content of the seeds was determined at the time of collection, after which portions of the seed were exposed to temperatures of 33°, 20° and −20°F. for 10 hours, respectively. Seed germination tests were made with each lot. The data obtained indicated that sorghum seed with a moisture content of 30 to 40 percent was not seriously reduced in vitality by an exposure to 20°F., but at −20°F. the damage was severe. When the moisture content was reduced to about 13 to 15

percent there was no injury by exposure to a temperature of −20°F.

Soybean seed with a moisture content of 50 to 63 percent withstood a temperature of 20°F. but was seriously damaged when the temperature reached −20°F. When the moisture content had reached 30 percent there was no appreciable injury to germination from a temperature of −20°F.

Timothy seed obtained by (a) combining from standing plants, and (b) threshing from the shock in 1940 was examined at the Iowa Station (2) for percentage of hulless seeds, and the germination of both hulless and unhulled seeds was determined. A summary of the results is given in table 2.

TABLE 2. GERMINATION OF COMBINED AND THRESHED TIMOTHY SEED.

Method of harvesting	No. lots	Percentage		Total pure seed	Percentage germination		
		Hulless	Unhulled		Hulless	Unhulled	Mixed
Threshed.....	9	29.3	68.3	97.6	91	96	95
Combined....	8	58.0	38.0	96.0	70	96	78

The data show that for the 1940 crop the percentage of hulless seeds was much higher and their vitality much lower in combined than in threshed seed.

Storage conditions and the percentage of moisture carried by the seed have a marked effect on the vitality of seeds (45). Data from the U. S. Department of Agriculture (3) show that although there is considerable variation in the ability of different kinds of vegetable crop seeds to retain their vitality at a high temperature and a high humidity, yet all crop seeds retain their viability longer when dried and stored at a temperature of 50°F. or less and in a relative humidity of not over 50 to 60 percent. Seed viability may be maintained longer if mature, dry seeds are stored in a sealed container at a temperature below 15° C.

Longevity of seeds of field crops stored in a laboratory or a granary varies by crops. Information from several sources concerning the length of life of stored seeds may be summarized briefly as follows:

(1) At Fort Collins, Colo. (44) seeds of several crops stored in a dry, unheated room for 10 years were sampled each year and tested for germination. The data showed that:
   (a) The germination of unhulled barley seed decreased about 14 percent in 10 years and naked barley lost vitality more rapidly than unhulled seed.
   (b) The germination of wheat seed decreased about 7 percent in 10 years.
   (c) The germination of oats decreased about 13 percent in 10 years.

(d) The germination of rosen rye and Wisconsin black soybeans declined 10 percent in 5 years and more rapidly thereafter.
(e) A decline of 2 percent in the germination of black amber sorghum seed occurred in 6 years.
(f) Yellow dent corn retained its vitality well for 4 years, but the germination decreased 13 percent the fifth year and 20 percent after 8 years of storage.

(2) In Canada (48) seeds of several crops were stored for 20 years, and germination tests were made each year. The collections were made in all parts of Canada and were obtained directly from farmers.
The samples included:
(a) Wheat—170 collections from 14 varieties
(b) Oats—179 collections from 30 varieties
(c) Timothy—25 samples
(d) Alsike and red clover—24 samples of each
The data from all tests showed that:
(a) Wheat seed retained its germinating power well for 5 years after which the weaker seeds died. At the end of 15 years about 75 percent of the seeds were no longer viable.
(b) The longevity of oat seed was longer than that of wheat. The seeds retained their vitality well for 10 years, then declined in vitality at the end of 15 years to about 80 percent of the original test.
(c) Timothy seed declined gradually in vitality the first 5 years, showing about 10 percent reduction in that time. At the end of the seventh year its vitality had declined 16 percent, and by the twelfth year the average germination was only 11.5 percent.
(d) Alsike and red clover seed declined gradually in viability; each germinated about 80 percent at the end of 4 years compared with 97 percent when collected. At the end of 7 years the germination was about 65 percent, and at 10 years about 45 percent.

(3) In the Iowa State College Seed Laboratory seeds of barley and sorghum showed relatively little loss in germination after storage in a dry heated room for 8 years, and cucumber seed decreased in germination only slightly after 7 years of laboratory storage. Viability of sweet corn seed declined about 10 percent in 8 years.

It should not be assumed that seed longevity of large lots stored in a warehouse or granary will be the same as that of similar seeds stored in small quantities in a dry laboratory room. If field crop seeds are well matured and dry when put in storage and if the storage room is dry and the relative hu-

midity reasonably low, the vitality of large lots will not be greatly different from that of small lots. In sub-tropical climates where humidity is high and temperatures fairly high throughout the year, seeds decline rapidly in viability (1). The safest practice is always to make tests before planting.

Genetic make-up affects seed vitality. In the development of hybrid seed corn it has been found that inbred lines vary greatly in the longevity of the seed. Some lines possess inherited factors that either hasten the decline or prolong the vitality of the seeds. It is evident, therefore, that seeds of lines differing in their inheritance may be expected to vary in longevity when stored under the same conditions.

Organisms carried by seeds may be responsible for a rapid decline in seed viability. Seeds of corn infected with *Diplodia zeae* and barley kernels infected with *Gibberella saubinetii* lose their vitality more rapidly than seeds not infected, even though the organisms may die before the infected embryos do. Seed disinfection with chemical dusts will often prolong the life of diseased seeds, although over-doses of volatile mercury compounds cause mercury poisoning and hasten the loss of seed vitality.

Seed corn treated with Merko or Semesan Jr. and kept in a dry room for 3 years germinated 95 percent at the end of the test. Seed of wheat, oats and barley treated with New Improved Ceresan (5 percent ethyl mercury phosphate) at the rate of ½ ounce to the bushel germinated over 90 percent after 3 years of dry storage.

Weevils, chalcis fly, grain moths and other insects commonly feed upon stored seed. In time, part or all of the infested seed may be consumed leaving only the edosperm or the seed coat.

SAMPLING, PACKAGING AND MAILING OF SEED

The working sample used in a seed laboratory is necessarily small, hence it should be as representative as possible. If the sample drawn from a seed lot is not representative then any test may be misleading. Large lots of seed usually consist of several small lots of a given kind or variety blended together, each grown on a different farm. If blending is careless or incomplete it is difficult to draw a representative sample.

SAMPLING SEED

Suggestions for sampling seeds preparatory to submission for analysis are as follows:

1. For lots of 10 bags or less, take one or two samples from each bag, either with an approved probe or by hand. Samples should be obtained from near the top, in the center and as near the bottom as possible. Mix all the

small samples together thoroughly and then subdivide until a smaller portion is obtained for the test sample.

2. For lots of over 10 bags, draw samples from every second, third, fifth or tenth bag, depending on the size of the lot, mix thoroughly together and then draw a sample for test. This may be done by drawing a portion from different parts of the mixed sample or by pouring it on to a table and dividing it into fourths, eighths, or smaller fractions.
3. Seed grain in bins may be sampled with a trier or probe. Several small samples may be drawn and mixed together as described above. Samples from a bin may also be taken in different parts and at different depths. The larger the number and wider the distribution of small samples that are taken the more representative will be the sample submitted for analysis.

AMOUNT OF SEED NEEDED

The amount of seed needed for a test sample varies with the size of the seed. The following amounts are preferred:

Timothy, bluegrass, clovers, alfalfa and other seeds of similar size, 4 ounces.

Flax, rape, sudan grass, sorghum, bromegrass and others of similar size, 8 ounces.

Small grains, soybeans and corn, 1 pound.

If a pearling or disease test is to be made on barley, at least 2 quarts of seed should be submitted.

PACKAGING AND MAILING

Cloth bags or tin boxes are preferable containers for seeds to be sent by mail, but heavy paper seed packets are used by seedsmen and are satisfactory. *Regular letter envelopes are unsatisfactory because they permit loss of seed.* The outside of the container should show the sender's name and address in legible letters and a slip or tag giving the kind of seed, the name and address of the sender should be placed inside the container. *Address the package to the Iowa State College Seed Laboratory, Ames, Iowa.* A letter of explanation attached to the package should be included.

METHODS EMPLOYED IN A SEED LABORATORY TO DETERMINE SEED QUALITY

Seeds are analyzed for purity and noxious weed seed content, tested for germination and examined for the presence of seed-borne organisms. The methods employed are numerous and require not only extensive equipment but highly technical skill on the part of seed analysts.

Fig. 1. Boerner sampler.

PURITY ANALYSIS

The size of sample used in a seed laboratory for a purity analysis or a noxious weed examination depends upon the kind of seed and the approximate number of seeds per gram, ounce or pound. Detailed purity analyses are time-consuming and require much patience and energy on the part of an analyst. It is practically impossible to use large samples, and the smaller the size of individual seeds the smaller the sample used. Rules for seed testing formulated by the Association of Official Seed Analysts and by the United States Department of Agriculture (46) for the enforcement of the Federal Seed Act provide for a laboratory working sub-sample that contains approximately 3,000 seeds, but there are variations from this approved standard. Whatever the weight of sub-sample used it is important that it be as truly representative of the bulk sample as possible. To obtain such a sub-sample it is necessary to pass the bulk sample through a divider or mixing machine which either subdivides or permits drawing small fractions from the mixer. The Boerner sampler (fig. 1) is widely used, and a mixer of the type illustrated in fig. 2 is satisfactory for chaffy grasses such as bromegrass, orchard grass, Rhode's grass and bluegrass. Table 3 shows the recommended size of sample for both purity analysis and noxious weed examination for many kinds of crop seeds. Included in the table is the approximate number of seeds per gram, ounce and pound of pure seed.

When the working sample is obtained it is not necessary to have the exact weight as given in table 3. It is better to use a sample obtained by the divider or sampler, even though it be a trifle more or less than the recommended weight, than it is to attempt to add or subtract from the divided portion by hand.

TABLE 3. APPROXIMATE WEIGHT IN GRAMS OF SAMPLES FOR PURITY ANALYSIS AND NUMBER SEEDS PER UNIT WEIGHT.

Kind of seed	Minimum weight for working sample	Approximate number seeds per unit weight of pure seed			Weight for noxious weed check (grams)†
		Gram	Ounce	Pound	
Alfalfa	5	500	14,170	226,720	50
Barley	100	30	850	13,600	500
Beet	50	54	1,530	24,480	300
Bentgrass	0.5*	18,000	510,300	8,164,800	25
Bluegrass					
Canada	1	5,500	155,920	2,494,720	25
Kentucky	1	4,800	136,000	2,150,080	25
Rough	1	5,600	158,760	2,556,160	25
Bromegrass	5*	300	8,500	136,000	50
Buckwheat	50	45	1,275	17,200	300
Carrot‡	2	900	25,510	408,160	50
Clovers					
Alsike	2	1,500	42,520	680,320	50
Alyce‡	5	680	19,260	308,160	50
Crimson	10	1,330	9,350	149,600	50
Hop‡	2	2,200	62,330	997,280	50
Persian‡	2	1,435	40,650	650,400	50
Red	5	600	17,010	272,160	50
Sweet	5	570	16,160	258,400	50
White	2	1,500	42,520	680,320	50
Dogstail	2	1,900	53,860	861,760	50
Fescue					
Chewings	2	1,200	34,000	542,720	50
Fine-leaved	2	1,200	34,000	542,720	50
Meadow	5	500	14,170	226,800	50
Red	2	755	21,400	342,400	50
Flax[1]	10	300	8,500	136 000	50
Grass					
Bahia	5	300	8,500	136,000	50
Bermuda	1	3,940	111,700	1,769,760	25
Carpet‡	1	2,930	83,000	1,328,000	25
Dallis‡	2	590	16,710	267,360	50
Johnson‡	15	270	7,650	122,400	150
Orchard	2	1,100	32,600	521,600	50
Reed Canary‡	2	1,500	42,520	680,320	50
Sudan	25	120	3,400	54,240	150
Lespedeza					
Chinese—hulless	5	820	23,250	372,000	50
Common—unhulled	5	750	21,260	340,160	50
Korean—unhulled	5	525	14,880	238,080	50
Millet					
Broom corn	25	180	5,100	81,600	150
Brown top‡	10	330	9,350	149,600	50
Cattail‡	10	170	4,800	76,800	50
Foxtail	5	470	13,320	213,120	50
Japanese‡	5	310	8,780	140,480	50
Oatgrass, tall	10	330	9,350	149,600	50
Oats	100	28	790	12,640	500
Rape—winter	10	230	6,520	104,320	50
Redtop	.5*	11,000	311,850	4,989,600	25
Rye	100	40	1,130	18,080	500
Ryegrass					
Italian	5	500	14,170	226,720	50
Perennial	5	500	14,170	226,720	50
Short-seeded perennial	5	700	19,840	317,440	50
Sorghum					
Amber	50	55	1,560	24,960	300
Atlas	50	55	1,560	24,960	300
Soybean	100	7	198	3,168	500
Timothy	2	2,500	70,870	1,133,920	50
Turnip	10	340	9,640	154,240	50
Vetch					
Common	100	19	538	8,600	500
Hairy	100	36	1,000	16,000	500
Wheatgrass					
Crested, fairway	5*	714	20,250	324,000	50
Crested, standard	10	425	12,050	192,800	50
Slender	10	340	9,640	154,240	50
Western	10	250	7,080	113,280	50
Wheat	100	25	708	11,328	500

* Weight for purity less than recommended in rules. Amount given here is based on experimental data.

† One ounce equals 28⅓ grams.

‡ Determined in Iowa State College Seed Laboratory, Ames, Iowa.

[1] Many new varieties have less than 200 seeds per gram.

Fig. 2. Iowa seed mixer and treater.

Weighing the working sample is important and should be to four significant figures. For example, samples weighing less than 1 gram should be weighed to the fourth decimal place, those ranging between 1.000 and 9.999 grams should be weighed to the third decimal place and those weighing between 10 and 99.99 grams to the second decimal place.

PURE SEED

The rules for seed testing adopted by the Association of Official Seed Analysts and the United States Department of Agriculture for the enforcement of the Federal Seed Act include as "pure seed" all seeds of the kind being tested whether shriveled, immature, insect injured, embryoless, discolored or broken so long as over half the seed is present. Seeds of clovers, alfalfa, crucifers and flax without the seed coats are not classed as pure seed, nor are seed hulls, chaff, empty grass florets, nematode galls of wheat and other grasses and fungous bodies such as ergot sclerotia and smut masses.

In practice the application of this definition is difficult and has not resulted in the degree of uniformity among seed laboratories that should be expected by the nature of the material or that should exist for the orderly marketing of seeds. A detailed exposition of this problem is given by Porter (31) and Porter and Leggatt (35).

The object of testing seed is to determine its seeding value. This is usually expressed in terms of purity and germination which may be further expressed as pounds of pure, viable seed per hundred. It is obtained by the formula

$$\frac{\text{percent pure} \times \text{percent germination.}}{100}$$

For example, if a seed lot has a purity of 96 percent and germination of 90 percent, the index value is $\frac{96 \times 90}{100} = 86.4$, or 86.4 pounds of pure viable seed out of each 100 pounds of the lot. This method of computing index value is accurate only when the unit weights of the

viable and non-viable particles in the pure seed fraction are the same. The reason for this is that pure seed is determined by weight and the germination percentage by number. The presence in the pure seed fraction of a considerable number of immature or undeveloped and non-viable structures that resemble seeds but that are lighter in weight than true seeds of the kind being tested, reduces the percentage of germination disproportionately to their percentage by weight in the pure seed fraction. An illustration of this relationship is provided by the data obtained by the Iowa Agricultural Experiment Station from the analyses of five lots of red clover infested with chalcis fly. Two methods of analysis were used (1) the official by which all immature, undeveloped and chalcis fly-infested seeds were retained in the pure seed fraction and (2) a modified procedure by which empty, undeveloped and chalcis fly-infested seeds were placed in the inert fraction. The following data show the percentages of pure seed and of germination together with the index values:

Sample no.	Official method			Modified method		
	Percent pure	Percent germination	Index value	Percent pure	Percent germination	Index value
Red Clover No. 1....	96.5	64.5	62.3	84.3	88.3	74.4
Red Clover No. 2....	99.5	81.5	81.1	95.0	94.0	89.3
Red Clover No. 3....	99.5	92.0	91.6	98.1	92.8	91.0
Red Clover No. 4....	96.3	87.0	83.8	95.2	90.5	86.2
Red Clover No. 5....	99.0	75.8	75.0	96.9	83.3	80.7
Average.....	.....	.....	78.8	.....	.....	84.1

Note that the index value (pounds per hundred of pure live seed) is much higher for samples 1, 2, 4 and 5 when the modified method of analysis was used. This indicates that the retention in the pure seed fraction of empty or undeveloped seeds will not give the true seeding value of the sample in terms of pure live seed.

On the other hand, the removal of empty, undeveloped and immature seeds must be done in a uniform manner without which uniform results from purity analyses cannot be expected. Furthermore, all germination tests should be made with pure seed, and if a lot of seed contains many empty hulls, sterile florets or many undeveloped seeds which are not removed in a standard, uniform manner, then the pure seed fractions cannot be uniform and percentages of germination from replicate fractions will differ greatly.

An illustration of the importance of providing uniform pure seed fractions is given by tests made in the Iowa State College Seed Laboratory with replicate sub-samples from a lot of Kentucky bluegrass seed. There were 64 sub-samples drawn and analyzed by a uniform method. The mean percentage of pure seed

was 88.64 and the range was 87.8 to 89.6. The mean percentage of germination was 88.5 as shown by testing 4 × 100 seeds from each of several pure seed fractions. The index value was thus found to be 78.45. To each of 32 laboratories a subsample, previously analyzed but recombined, was sent for analysis. The range in purity from 26 reports was 87.4 to 94.2, and the range in germination was 55 to 91. The laboratory that obtained 94.2 percent purity reported 55 percent germination which gave an index value of 51.81, while the highest value obtained was 82.17. It is apparent that the pure seed fraction from the sample which had a high percentage of pure seed contained many empty or immature particles, light in weight and incapable of germination, hence the index value obtained was not accurate for the lot. There is now conclusive evidence that purity percentages based on the presence of light weight, immature and undeveloped structures in the pure seed fractions are not compensated for in the germination result.

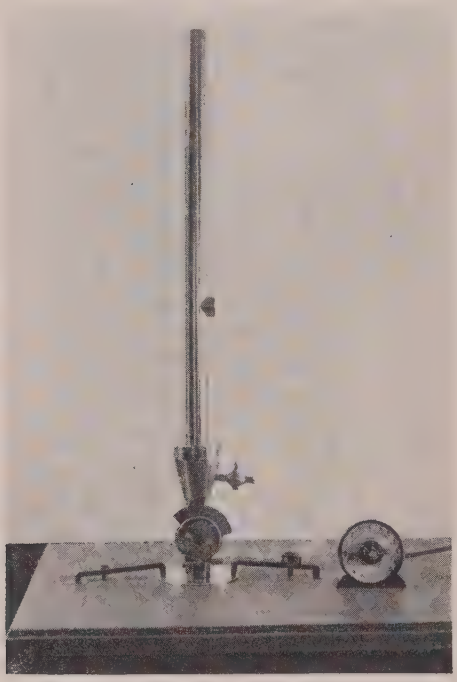

Fig. 3. Iowa air blast seed separator.

The use of an air blast seed separator such as illustrated in fig. 3 and equipped with a device for critical control of air pressure is essential to the uniform analysis of many kinds of seeds, particularly those which contain structures that are empty but cannot be differentiated readily from normally developed seeds. A larger unit of the same type is used for sudan grass, bromegrass, wheat grasses and cereals.

OTHER CROP SEED

In this class are placed all seeds from plants classed as a crop if the amount does not exceed 5 percent by weight of the sample. The same rule as to broken, insect-injured and immature seed applies as given for pure seed. When any one kind of crop seed exceeds 5 percent by weight the lot is considered a mixture.

WEED SEED

All seeds of plants commonly classed as weeds, including noxious weeds, are placed in this class. Shriveled, immature or injured weed seeds clearly incapable of germination are placed in the inert fraction, and the paragraph describing inert matter includes a list of specific types which should be classed as inert material. When seeds of path rush (*Juncus tenuis*) are present in an amount that would not add more than 0.1 percent to the percentage of weed seed, they need not be separated with the weed seed but may be included with the inert matter. However, the presence of such seeds should be recorded.

INERT MATTER

The inert fraction includes chaff, empty glumes and attached sterile florets of grasses, dirt, stones, fungous bodies (ergot, smut balls), broken or insect-injured crop seeds half or less in size and such crop seeds as rape, cabbage and other crucifers, legumes and flax when the seed coat is absent. In addition the inert fraction includes undeveloped and badly injured weed seeds that by visual examination are considered incapable of germination.

The term "visual examination" is an unfortunate one to employ in the rules for seed testing. It implies that which can be perceived by vision and therefore could be interpreted to mean those structures which by external appearance are plainly undeveloped and could not grow. It could also be interpreted to mean those structures which when dissected may be perceived as plainly empty and devoid of a seed embryo.

Repeated observations and tests by the writer have shown that in such families as Gramineae, Polygonaceae, Cyperaceae, Convolvulaceae and Compositae, undeveloped structures which by external appearance resemble well developed seeds or fruits are common and occur frequently in commercial seed samples of crop plants. For example, empty fruit cases of (a) sheep sorrel and sedge occur in bluegrass seed, (b) sour dock occur in orchard grass, and (c) Canada thistle and chicory occur in legume seed. Empty seed cases of dodder occur in clover and lespedeza seed and of bindweed in small grain seed. Such structures have too long been classed as weed seeds by seed analysts. Many of our seed crops are harvested at a time when certain weed plants have not developed sufficiently to produce mature, viable seed. Other

crop plants may have such plants as Canada thistle growing among them, yet the dioecious character of the Canada thistle (*Cirsium arvense*) is such that only a small percentage of its flowers are ever fertilized. Insects frequently infest weed seeds and consume part or all of the embryo.

Unfortunately in the cleaning process of some grass and legume seed lots it is impossible to remove all the empty and immature weed seeds without removing a considerable amount of good crop seed. This fact is evidenced not only by the presence of such structures in crop seed of good quality but also by tests with controlled air pressure in the Iowa Seed Laboratory which reveal that a pressure sufficient to remove all empty or undeveloped weed seed hulls will remove many pure seeds of bluegrass and orchard grass. The reverse is also true. In carpet grass tests many well developed fruits of *Fimbristylis sp.* are removed with the empty grass florets.

Detection of empty or undeveloped structures that resemble weed seeds or fruits is impossible by visual examination of external features alone. It is necessary to employ tweezers or a scalpel. Experience with an air blast seed separator in which air pressure can be controlled is of great value in separating firm, well developed and true seeds or fruits of weed plants from false seeds or fruits. In conjunction with a binocular or a lens, a scalpel or a pair of tweezers it is possible to make an arbitrary yet reasonably accurate separation. This procedure is necessary if seed testing is to be maintained on a scientific and practical level.

There are instances, however, in which external appearance may be relied upon to make reasonably accurate separation. Certain buckhorn seeds (39) have been shown to be non-viable, and ashen-colored structures (29) from dodder plants that resemble seeds have been found to be infertile and non-viable. Several investigators in reports of the Research Committee of the Association of Official Seed Analysts and in the News Letter have shown that certain types of weed structures are non-viable.

These recent efforts to classify weed seeds into two general classes marks a change in concept almost as marked as the change in concept (35) of what constitutes pure seed. Wright was one of the first to make tests of injured weed seeds and in a report to the International Seed Testing Association (54) proposed that weed seeds should be classed as inert matter when they are completely crushed or when the embryo is visibly missing. It now seems evident that a clearer definition of a seed from the seed laboratory point of view is necessary. The writer believes that insofar as possible, in seed laboratory practice, the term seed should mean a seed, a fruit or a multiple fruit (beet) in the strictest botanical sense (with one or more ripened ovules)

and that it should apply to either weeds or crops. Acceptance of that point of view would immediately result in the exclusion, from the category of either crop or weed seed, of all structures that never developed an embryo or that have a poorly developed embryo. Broken seeds and embryoless seeds present a special problem. Embryoless seeds of crop plants in the grass family should probably be retained as part of the pure seed fraction although placement of such particles and of plainly broken seeds of all crop plants in the inert fraction would give an index value much more accurate than the present rule provides (29).

Experimental evidence from many published reports of the Association of Official Seed Analysts and from the citations already referred to indicates that the following may be considered inert material.

(a) Embryoless fruits of all weedy grasses such as *Chaetochloa spp., Agropyron repens, Bromus secalinus* and *Bromus tectorum.* If over half the embryo is absent the seed cannot produce a normal seedling and it properly belongs in the inert fraction;

(b) Infertile and empty structures from dodder plants (*Cuscuta spp.*) which resemble seeds but which are usually fragile, ashen-grey to brown in color, somewhat enlarged and without an embryo;

(c) Ragweed seeds (*Ambrosia artemaesifolia*) with both the involucre and pericarp absent;

(d) Shriveled, blackened seeds of Buckhorn (*Plantago lanceolata*);

(e) Empty or immature fruits (seeds) of the Cyperaceae, Polygonaceae, Convolvulaceae and Compositae families resulting either from failure of the embryo to develop or from insect damage; such structures occur commonly among fruits of Canada thistle, sedge, sour dock and sheep sorrel and among seeds of dodder.

(f) Empty glumes and steril florets of weedy grasses;

(g) Naked seeds (seed coats absent) of the species of *Brassica* usually considered as weeds and of Leguminosae.

(h) All other crushed or broken weed seeds which appear clearly incapable of germination;

(i) Bulblets of wild onion and garlic with the basal or stem-end portion removed.

An air blast seed separator will aid greatly in separating all empty and some sterile structures from normal ones in classes (b), (e) and (f).

CALCULATION OF PERCENTAGES

Calculations to determine percentages of pure seed, inert, weeds and other crop seeds are made by dividing the total final

weight into each of the respective weights of the separations. An illustration of the procedure follows:

Red clover test	Weight in grams	Percent by weight
Sample before analysis	5.028	.....
Pure seed after analysis	4.881	97.19
Other crop seeds after analysis	.030	.60
Inert after analysis	.088	1.75
Weed seeds after analysis	.023	.46
Total after analysis	5.022	100.00

$$\frac{4.881}{5.022} = .9719 \times 100 = 97.19 \text{ per cent}$$

$$\frac{.030}{5.022} = .0060 \times 100 = 0.60 \text{ per cent}$$

$$\frac{.088}{5.022} = .0175 \times 100 = 1.75 \text{ per cent}$$

$$\frac{.023}{5.022} = .0046 \times 100 = 0.46 \text{ per cent}$$

The reduction in weight from 5.028 to 5.022 grams may have been caused by loss in moisture or loss of a few particles. Reduction in moisture is quite common and sometimes amounts to 5 percent. In some instances moisture absorption is pronounced, depending on the humidity and temperature of the atmosphere and the condition of the seed.

SEED IDENTIFICATION

One of the most important phases of the purity analysis is the identification of all the seeds; otherwise it is often impossible to place a seed in the weed or crop class. Designation as a weed or a crop is not always an arbitrary matter because some plants classed as weeds 10 or more years ago have come to be used as crops in certain areas or for a specific purpose. Nevertheless, a list of plants generally classed as crops is necessary, and reference to tables 3 and 4 will assist analysts in seed classification.

The methods employed in seed identification are as follows:

(a) Examination of external features. By experience it is usually possible to place a seed in the proper family by external appearance alone. For example, naked grass fruits have a small yet readily distinguishable embryo partially enclosed by a relatively large endosperm. The hilum and embryo are always on the same end but opposite each other. There is no other plant family whose seeds (or fruits) have such external features. Grass fruits (of commerce) may also be enclosed by a lemma

and palea, the palea being partially or completely covered by the lemma. A large number of grass fruits have a small stalk (rachilla) attached near the base of the glumes on the palea side. The shape, length and degree of pubescence of the rachilla aid in identification. The oat grain shown in fig. 4, A 1, 2, 3 and 4 illustrates the several structures to which reference has been made. Seeds of the dock or buckwheat family are really one-seeded fruits covered with a pericarp (ovary wall) and sometimes with an extra covering (old flower calyx) over the pericarp. The fruits in this family are either flattened or three-sided, usually brown to black in color and the pericarp is commonly lustrous (in some species not). The axis of the ovule in this family is straight and the micropyle is at the tip with the hilum at the opposite or basal end as shown in fig. 4, E-1.

Seeds of the pigweed and lamb's quarters' families are similar in shape and color and are characterized by a narrow embryo coiled around a mealy endosperm. (Fig. 4, H).

Seeds of the cockle family are commonly somewhat kidney shaped with the hilum near the center of the concave surface. The surface of the seed coat is usually rough with tubercles arranged either irregularly or in concentric rings. (Fig. 4, F.)

Seeds of the legume family vary greatly in size from tiny ones like the hop clover to the lima and the horse beans. The shape and color are equally variable. Heart, kidney, spherical and oval shapes are common. The color may be solid or mottled, and the seed coat surface is dull or lustrous. One characteristic feature of all legume seeds is that the strophiole and micropyle are close to but on opposite sides of the hilum. The raphe is also usually noticeable. These structures are illustrated in fig. 4, C.

Seeds of the mustard, morning-glory, mallow, vervain, mint, plantain and composite families have rather well defined characteristics that become evident to an analyst as experience is gained.

A seed herbarium with verified samples is essential to accurate identification of seeds. The seeds may be filed in small glass vials and arranged in a cabinet alphabetically within each family. It is also helpful to collect the plants, mount them on cardboard, place the seeds in a glassine bag and attach to the cardboard. This will encourage familiarity with both the plant and its seeds. The Seed Laboratory at Iowa State College will identify seed samples free of charge.

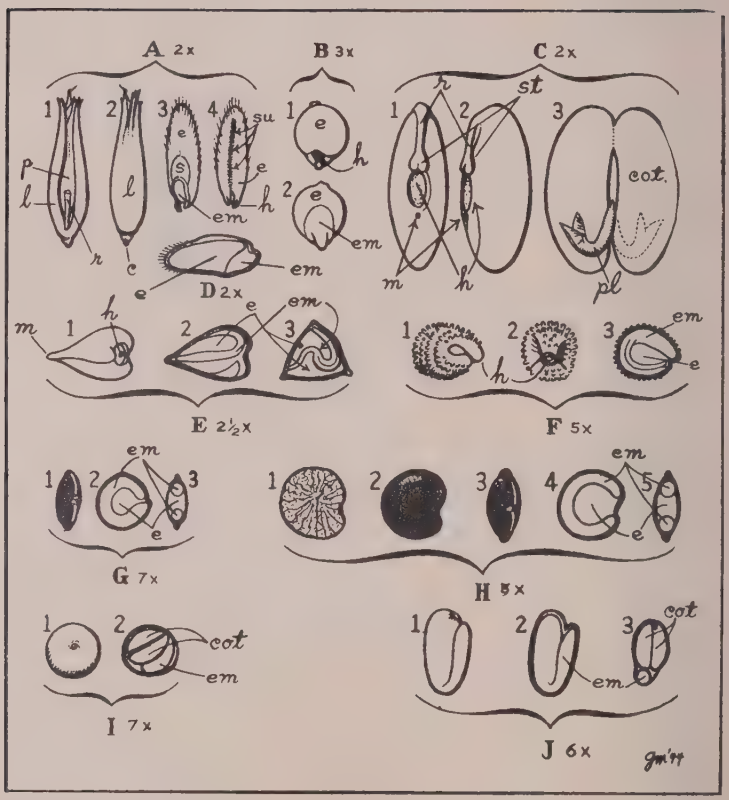

Fig. 4. Characteristic features of seed in different families.

A and B—Grass family.
- A1 —Oat grain with lemma (l) and palea (p) showing rachilla (r).
- A2 —Lemma view of oat grain showing callus (c).
- A3 —Dorsal view of naked oat grain showing embryo (em) scutellum (s) and endosperm (e).
- A4 —Ventral view of naked oat grain showing sulcus (su) and hilum (h).
- B1-2 —Ventral and dorsal views of sorghum seeds showing same features as in naked oat grain.
- C —Legume family.
- C1-2 Garden bean with raphe (r), hilum (h), strophiole (st) and micropyle (m).
- C3 —Garden bean split longitudically showing cotyledon (cot) and plumule (pl).
- D —Longitudinal section of wheat grain showing endosperm (e) and embryo (em).
- E —Buckwheat family.
- E1 —Seed of buckwheat with pericarp removed showing hilum (h) and micropyle (m).
- E2-3 —Longitudinal and transverse sections of buckwheat grain showing embryo (em) and endosperm (e).
- F —Cockle family.
- F1-2 - Exterior views of corn cockle (**Agrostemma githago**) seeds showing hilum (h).
- F3 —Internal view of longitudinal section of corn cockle seed showing relative positions of embryo (em) and endosperm (e).

G —Pigweed family.
G1 —Exterior view of seed of prostrate pigweed (**Amaranthus blitoides**) with hilum.
G2-3 —Longitudinal and transverse sections of pigweed seed with embryo (em) and endosperm (e).
H —Goosefoot family.
H1-2-3 —Exterior view of seeds of maple leaved goosefoot (**Chenopodium hybridum**).
H4-5 —Longitudinal and transverse sections of seed of maple leaved goosefoot with embryo (em) and endosperm (e).
I and J —Mustard family.
I1 —Exterior view of seed of wild mustard (**Brassica arvensis**).
I2 —Longitudinal section of mustard seed showing cotyledons (cot) and embryo (em).
J1 —Exterior view of seed of false flax (**Camelina sativa**).
J2-3 —Longitudinal and transverse sections of false flax seed showing embryo (em) and cotyledons (cot).

A fairly good substitute for a herbarium is a set of illustrations showing the external features of seeds. Figures 5 to 21 will be found of great value in seed identification. The smallest drawing outline in the illustration of each species represents approximately the natural size of the seeds. The botanical as well as the common name is given under each illustration and will be of value to those interested in seed taxonomy.

(b) Examination of internal structure.

The arrangement of the embryo with respect to the endosperm, its size and shape are features that are useful for family classification. A section perpendicular to the embryo axis of seeds of the goosefoot family will reveal the embryo region at the edges of the sections, and a section parallel to the axis will show the embryo coiled around the mealy endosperm (see fig. 4, H). Figure 4, A, B, D, E, F, G, I and J, illustrates the arrangement of the embryo and the cotyledons in seeds of the mustard, buckwheat, cockle, pigweed and grass families.

(c) Physico-chemical methods.

Light and chemical reactions play a limited part in seed identification. The most practical application of light is that of the identification of seedlings of *Lolium perenne* (perennial ryegrass) and *Lolium multiflorum* (annual ryegrass) by means of ultra-violet light (23). The roots of the latter when grown on white filter paper fluoresce (show violet) if placed under an ultra-violet lamp in a dark room. The roots of the perennial form do not.

The seedlings of wild white clover may be differentiated reasonably well from those of the common white Dutch clover by treating the seedlings with picric acid. The seedlings of the wild form contain a glucoside which hydrolyzes at 30° C. (86°F.) and liberates hydrocyanic acid gas which will react with filter paper previously treated with picric acid, thereby producing a brown color.

LIST OF SEEDS BY PLANT FAMILIES AND LOCATION OF MEMBERS OF EACH FAMILY BY PLATE AND NUMBER.

Family name		Figure no.	Specimen no.
Common	Botanical		
Grass	*Gramineae*	5, 6, 7	1 to 24 inclusive
		20	1 to 13 inclusive
		21	1, 2, 3
Sedge	*Cyperaceae*	8	1 to 9 inclusive
Rush	*Juncaceae*	8	10, 11, 12
Lily	*Liliaceae*	8	13
Nettle	*Urticaceae*	8	14
Buckwheat	*Polygonaceae*	8	15 to 24 inclusive
		9	1 and 2
Goosefoot	*Chenopodiaceae*	9	3 to 12 inclusive
		20	14
Pigweed	*Amaranthaceae*	9	13 and 14
Carpet-weed	*Aisoaceae*	9	15
Cockle	*Caryophyllaceae*	9	16 to 24 inclusive
		10	1 to 6 inclusive
Whitlow-wort	*Illecebraceae* (*Corrigiolaceae* of some manuals)	10	7
Crowfoot	*Ranunculaceae*	10	8 to 13 inclusive
Poppy	*Papaveraceae*	10	14 to 18 inclusive
Mustard	*Cruciferae* (*Brassicaceae* of some manuals)	10	19 to 24 inclusive
		11	1 to 24 inclusive
Mignonette	*Resedaceae*	12	1
Rose	*Rosaceae*	12	2 to 8 inclusive
Legume	*Leguminosae*	12	9 to 24 inclusive
		13	1 to 24 inclusive
		14	1 to 5 inclusive
		20	15 to 19 inclusive
		21	4 and 5
Wood-sorrel	*Oxalidaceae*	14	6
Flax	*Linaceae*	14	7 and 8
Geranium (Cranesbill)	*Geraniaceae*	14	9 to 14 inclusive
Caltrop	*Zygophyllaceae*	21	6
Spurge	*Euphorbiaceae*	14	15, 16 and 17
		20	20 and 21
Mallow	*Malvaceae*	14	18 to 23 inclusive
St. Johnswort	*Hypericaceae*	14	24
Violet	*Violaceae*	15	1
Loosestrife	*Lythraceae*	15	2
Evening primrose	*Onagraceae*	15	3 to 7 inclusive
Carrot	*Umbelliferae* (*Ammiaceae* of some manuals)	15	8 to 13 inclusive
Primrose	*Primulaceae*	15	14
Bindweed (morning-glory)	*Convolvulaceae*	15	15 to 22 inclusive
		21	7
Borage	*Boraginaceae*	15	23 and 24
		16	1, 2 and 3
Vervain	*Verbenaceae*	16	4 to 7 inclusive
Mint	*Labiatae*	16	8 to 24 inclusive
		17	1 to 4 inclusive
Nightshade	*Solanaceae*	17	5 to 8 inclusive
		20	22 and 23
		21	8 to 11 inclusive
Figwort	*Scrophulariaceae*	17	9 to 15 inclusive
Plantain	*Plantaginaceae*	17	16 to 20 inclusive
Madder	*Rubiaceae*	17	21 to 24 inclusive
Valerian	*Valerinaceae*	18	1, 2 and 3
Teasel	*Dipsacaceae*	18	4 and 5
Bluebell	*Campanulaceae*	18	6
Lobelia	*Lobeliaceae*	18	7
Composite	*Compositae*	18	8 to 24 inclusive
		19	1 to 24 inclusive
		20	24
		21	12 to 16 inclusive

1
Sorghum vulgare sudanense
Sudan grass
2
Sorghum halepense
Johnson grass
3
Paspalum setaceum
4
5
Paspalum dilatatum
6
Digitaria sanguinalis
Crabgrass
7
Digitaria ischaemum
Smooth crabgrass
8
Panicum capillare
9
Panicum dichotomiflorum
Fall panicum
10
Panicum miliaceum
Proso, hog millet
11
Panicum virgatum
Switchgrass
12
Panicum sp.
No common name
13
Echinochloa crusgalli
Barnyard grass
14
Echinochloa crusgalli frumentacea
Japanese barnyard millet
15
Setaria lutescens
Yellow foxtail
16
Setaria viridis
Green foxtail
17
Setaria italica
Hungarian millet
18
Setaria italica
Common millet
19
Setaria italica
German millet
20
21
Dactylis glomerata
Orchard grass
22
Cynosurus cristatus
Crested dogtail
23
Aristida dichotoma
Poverty grass
24
Phleum pratense
Timothy

Fig. 5.

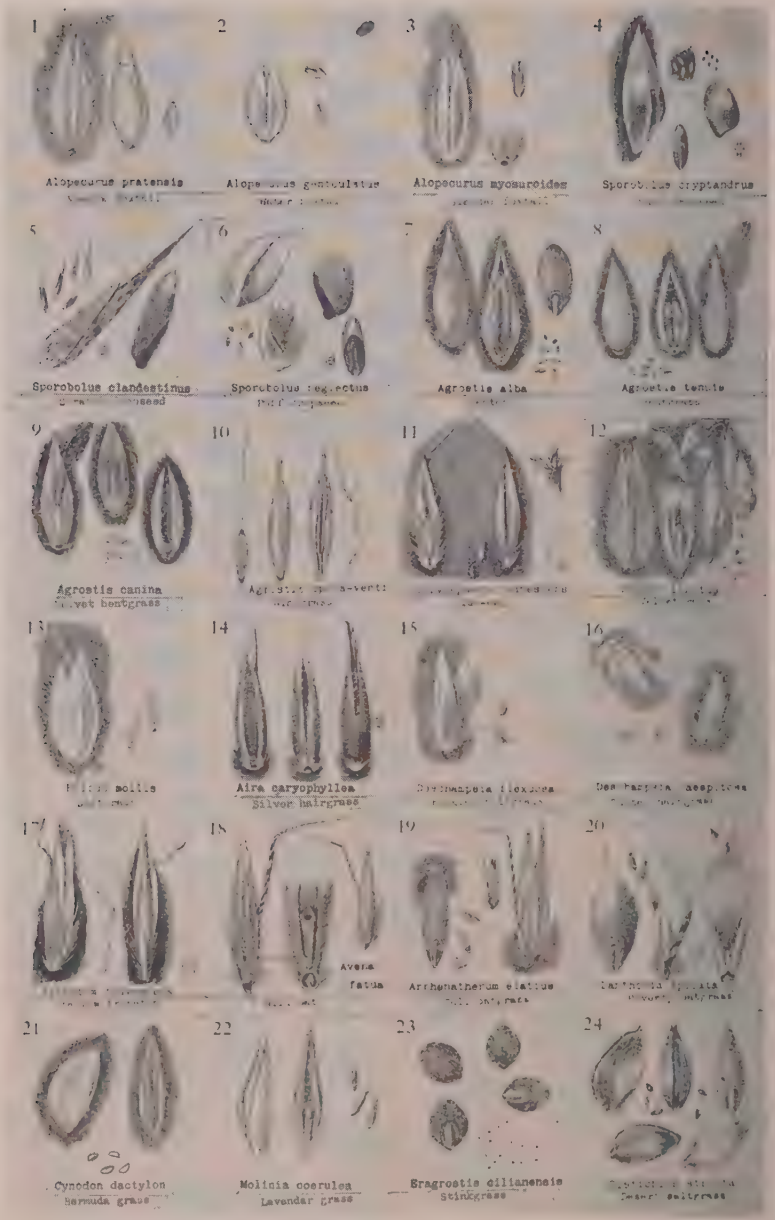
1
Alopecurus pratensis
2
3
Alopecurus myosuroides
4
Sporobolus cryptandrus
5
Sporobolus clandestinus
6
Sporobolus neglectus
7
Agrostis alba
8
9
Agrostis canina
velvet bentgrass
10
11
12
13
14
Aira caryophyllea
Silver hairgrass
15
16
17
18
Avena
fatua
19
Arrhenatherum elatius
20
21
Cynodon dactylon
Bermuda grass
22
Molinia coerulea
Lavendar grass
23
Eragrostis cilianensis
Stinkgrass
24

Fig. 6.

Fig. 1

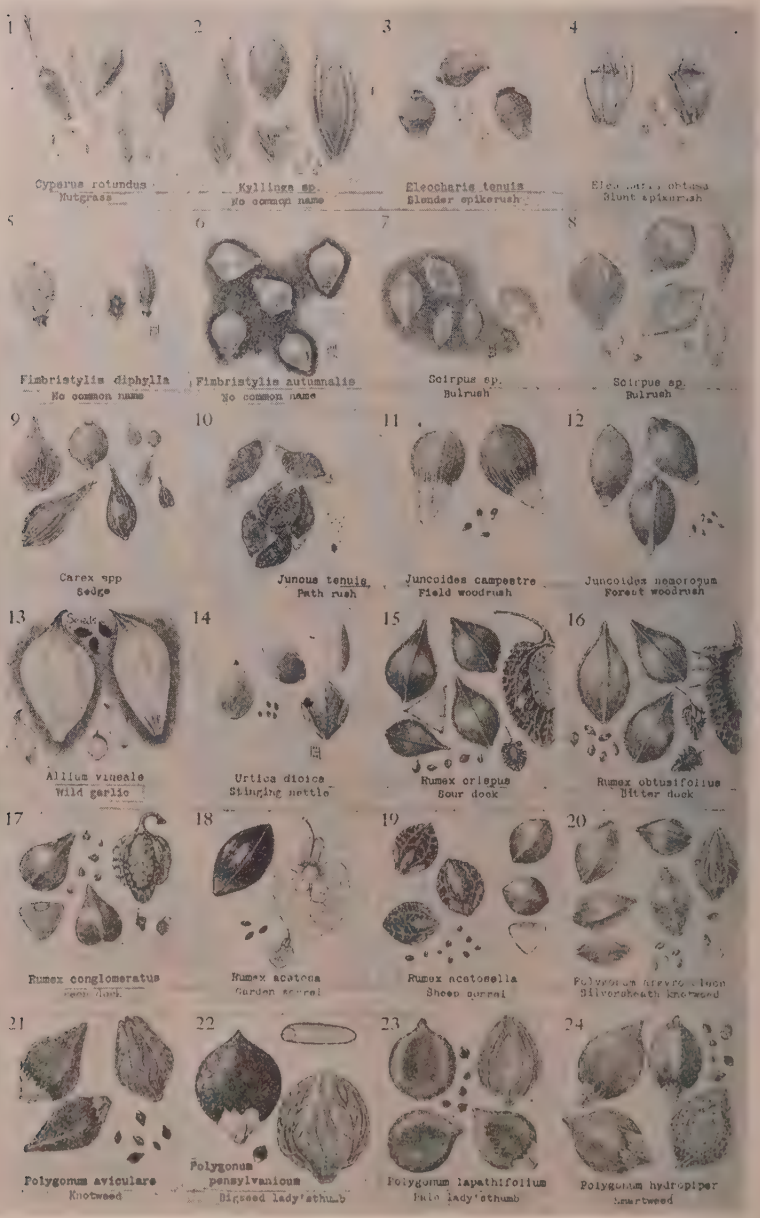
1
Cyperus rotundus
Nutgrass
2
Kyllinga sp.
No common name
3
Eleocharis tenuis
Slender spikerush
4
Eleocharis obtusa
Blunt spikerush
5
Fimbristylis diphylla
No common name
6
Fimbristylis autumnalis
No common name
7
Scirpus sp.
Bulrush
8
Scirpus sp.
Bulrush
9
Carex spp
Sedge
10
Juncus tenuis
Path rush
11
Juncoides campestre
Field woodrush
12
Juncoides nemorosum
Forest woodrush
13
Allium vineale
Wild garlic
14
Urtica dioica
Stinging nettle
15
Rumex crispus
Sour dock
16
Rumex obtusifolius
Bitter dock
17
Rumex conglomeratus
18
Rumex acetosa
Garden sorrel
19
Rumex acetosella
Sheep sorrel
20
Polygonum arenastrum
Silversheath knotweed
21
Polygonum aviculare
Knotweed
22
Polygonum pensylvanicum
Pigseed lady'sthumb
23
Polygonum lapathifolium
Pale lady'sthumb
24
Polygonum hydropiper
Smartweed

Fig. 8.

1
Polygonum persicaria
Lady's-thumb
2
Polygonum convolvulus
Wild buckwheat
3
Chenopodium album
Dock-pigeon's
4
Chenopodium berlandieri
Pitseed goosefoot
5
Chenopodium hybridum
Sharp-leaf goosefoot
6
Chenopodium murale
Nettle-leaf goosefoot
7
Atriplex hastata
Saltbush
8
Atriplex truncata
Orach
9
Bondia depressa
Swinepod
10
Salsola postifer
Russian thistle
11
Axyris amaranthoides
Russian pigweed
12
Bassia hyssopifolia
Bassia
13
Amaranthus retroflexus
Amaranthus graecizans
Redroot and Tumbling pigweed
14
Amaranthus blitoides
Prostrate pigweed
15
Trianthema portulacastrum
Horse-purslane
16
Agrostemma githago
Cornockle
17
Silene latifolia
Bladder campion
18
Silene dichotoma
Forked catchfly
19
Silene antirrhina
Sleepy catchfly
20
Silene noctiflora
Nightblooming catchfly
21
Silene anglica
English catchfly
22
Lychnis alba
White cockle
23
Vaccaria pyramidata
Cowcockle
24
Dianthus armeria
Deptford pink

Fig. 9.

1
Alsine media
2
Alsine graminea
3
Cerastium vulgatum
4
Arenaria serpyllifolia
Sandwort
5
Spergula arvensis
Corn spurry
6
7
8
9
Ranunculus acris
Tall buttercup
10
Ranunculus repens
Creeping buttercup
11
12
Ranunculus parviflorus
Crowfoot
13
14
Papaver somniferum
Opium poppy
15
Papaver
dubium
Papaver
rhoeas
16
Papaver sp.
Poppy
17
Papaver sp
Poppy
18
Glaucium corniculatum
Sea poppy
19
Lepidium virginicum
20
Lepidium densiflorum
Prairie peppergrass
21
Lepidium campestre
22
Lepidium draba
Perennial peppergrass
23
Hoary cress
24
Whitetop

Fig. 10.

1
2
3
4
5
6
7
8
9
10
Brassica arvensis
11
12
Camelina sativa
13
Camelina sativa
14
Bursa bursa pastoris
15
Arabidopsis thaliana
16
17
Camelina microcarpa
18
Neslia paniculata
19
Agrabis glabra
20
Cheiranthus cheiranthoides
21
Alyssum alyssoides
22
Berteroa incana
23
Eruca sativa
24

Fig. 11.

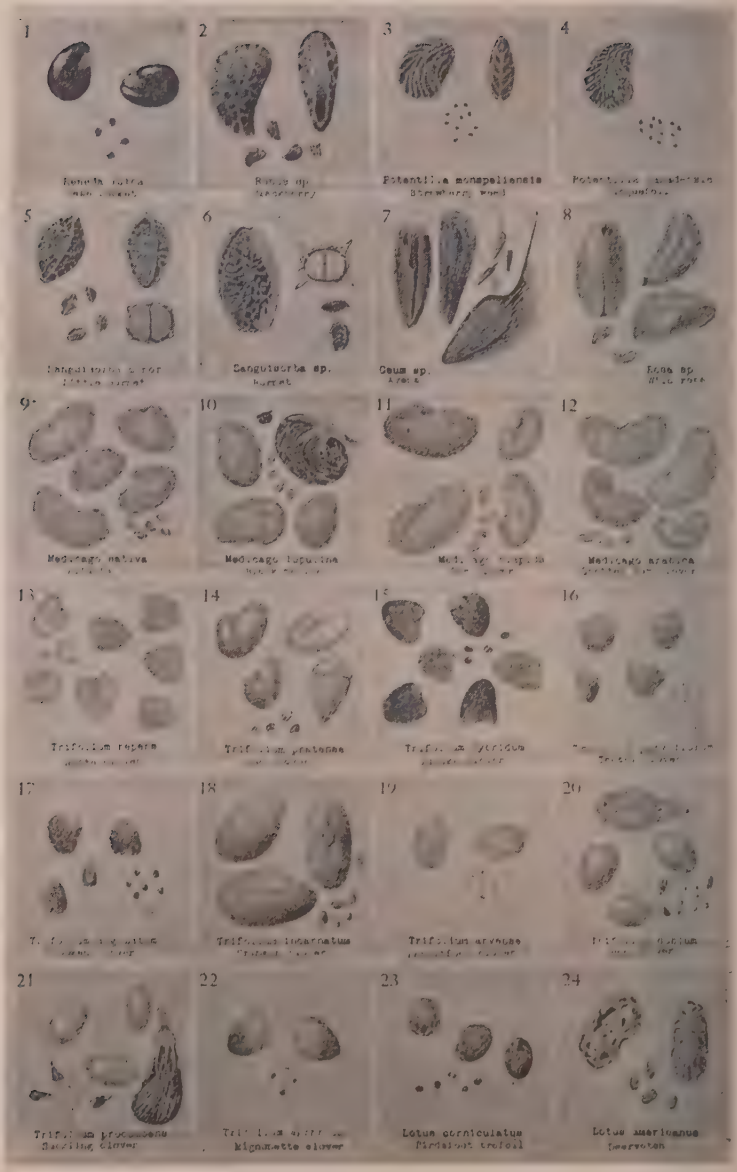
1
2
3
4
Potentilla monspeliensis
5
6
7
8
9
10
11
12
Medicago sativa
13
14
15
16
17
18
19
20
21
22
23
24
Lotus corniculatus
Birdsfoot trefoil
Lotus uliginosus

Fig. 12.

1
Melilotus alba
White sweet clover
2
Melilotus officinalis
Yellow sweet clover
3
Melilotus indica
Sour clover
4
Anthyllis vulneraria
Kidneyvetch
5
Coronilla varia
Crownvetch
6
Lespedeza striata
Common lespedeza
7
Vicia villosa
Hairy vetch
8
Vicia
angustifolia
Narrowleaf vetch
9
Vicia
angustifolia
Narrowleaf vetch
10
Vicia sativa
Common vetch
11
Vicia sativa
Common vetch
12
Vicia sativa
Pearl vetch
13
Vicia sp.
No common name
14
Vicia lutea
Yellow vetch
15
Vicia hirsuta
Tiny vetch
16
Vicia grandiflora
No common name
17
Vicia tetrasperma
Fourseed vetch
18
Vicia cracca
Bird vetch
19
Vicia gracilis
Slender vetch
20
Lathyrus
tuberosus
Groundnut pea
21
Lathyrus hirsutus
Rough pea
22
Lathyrus aphaca
Yellow pea
23
Pisum sativum arvense
Field pea
24
Garden pea

Fig. 18.

1
Trigonella foenum graecum
Fenugreek
2
Onobrychis viciaefolia
Sainfoin
3
Ornithopus sativus
Serradella
4
5
Alhagi pseudalhagi
Camelthorn
6
Oxalis stricta
Yellow oxalis
7
Linum usitatissimum
Flax
8
Linum virginianum
Woodland flax
9
Geranium dissectum
Cutleaf cranesbill
10
Geranium carolinianum
Carolina cranesbill
11
Geranium columbinum
Longstalk cranesbill
12
13
Geranium pusillum
14
Erodium cicutarium
15
Acalypha virginica
16
Euphorbia preslii
17
Euphorbia maculata
18
Sida spinosa
Prickly sida
19
20
Malva rotundifolia
21
Malva parviflora
22
Althaea hirsuta
23
Hibiscus trionum
Flower-of-an-hour
24
Hypericum perforatum
St. Johnswort

Fig. 14.

Fig. 24

Fig. 16.

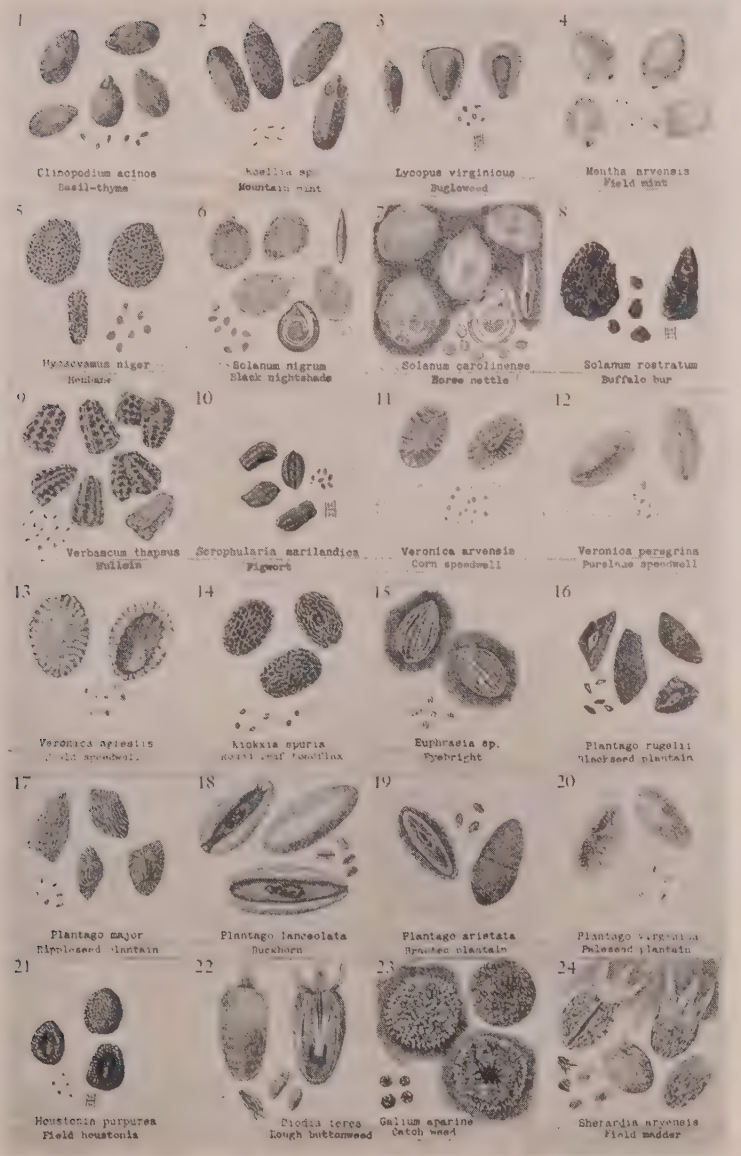
1
Clinopodium acinos
Basil-thyme
2
Koellia sp.
Mountain mint
3
Lycopus virginicus
Bugleweed
4
Mentha arvensis
Field mint
5
Hyoscyamus niger
Henbane
6
Solanum nigrum
Black nightshade
7
Solanum carolinense
Horse nettle
8
Solanum rostratum
Buffalo bur
9
Verbascum thapsus
Mullein
10
Scrophularia marilandica
Figwort
11
Veronica arvensis
Corn speedwell
12
Veronica peregrina
Purslane speedwell
13
Veronica agrestis
Field speedwell
14
Kickxia spuria
Round-leaf toadflax
15
Euphrasia sp.
Eyebright
16
Plantago rugelii
Blackseed plantain
17
Plantago major
Ribbleseed plantain
18
Plantago lanceolata
Buckhorn
19
Plantago aristata
Bracted plantain
20
Plantago virginica
Paleseed plantain
21
Houstonia purpurea
Field houstonia
22
Diodia teres
Rough buttonweed
23
Galium aparine
Catch weed
24
Sherardia arvensis
Field madder

Fig. 17.

Fig. 18.

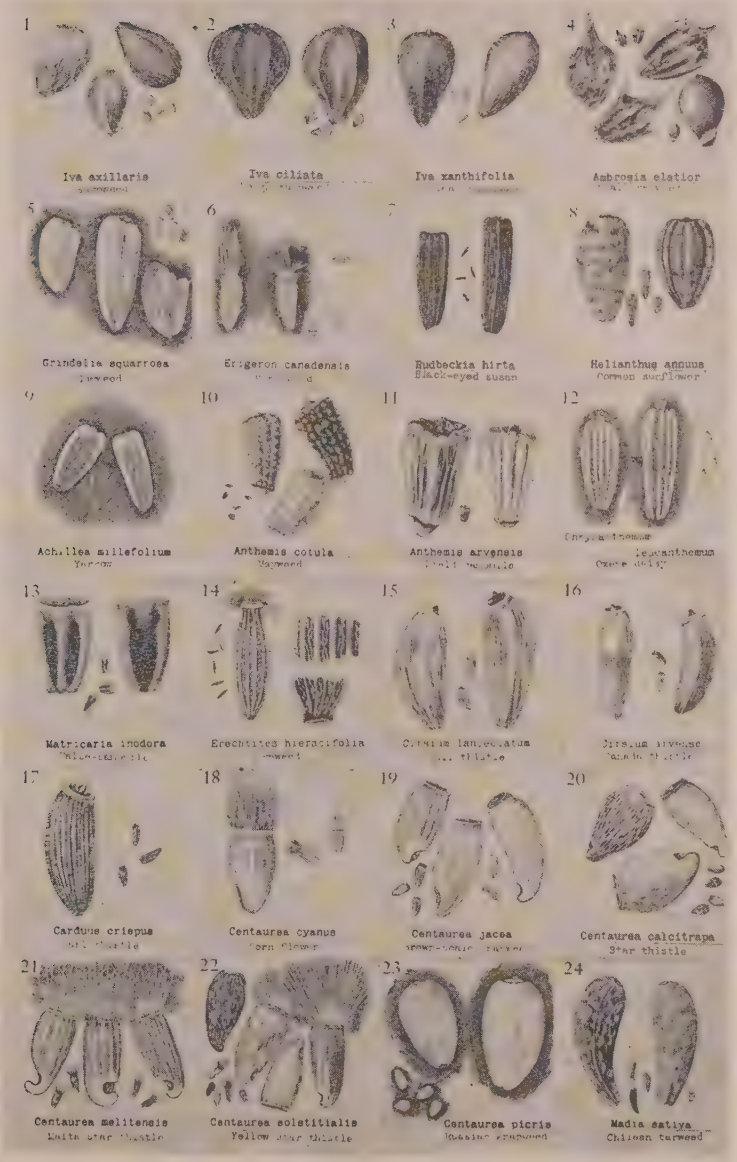
1
Iva axillaris
2
Iva ciliata
3
Iva xanthifolia
4
Ambrosia elatior
5
Grindelia squarrosa
Gumweed
6
Erigeron canadensis
7
Rudbeckia hirta
Black-eyed susan
8
Helianthus annuus
Common sunflower
9
Achillea millefolium
Yarrow
10
Anthemis cotula
Mayweed
11
Anthemis arvensis
12
Leucanthemum
Ox-eye daisy
13
Matricaria inodora
Wild chamomile
14
Erechtites hieraciifolia
15
Cirsium lanceolatum
16
Sonchus arvensis
17
Carduus crispus
18
Centaurea cyanus
Corn flower
19
Centaurea jacea
20
Centaurea calcitrapa
Star thistle
21
Centaurea melitensis
Malta star thistle
22
Centaurea solstitialis
Yellow star thistle
23
Centaurea picris
24
Madia sativa
Chilean tarweed

Fig. 19.

Bahia grass
2
Carrot grass
3
Panicum fasciculatum
Brown top millet
4
Phalaris arundinacea
Reed canary grass
Chloris gayana
Rhodes grass
6
Korean grass
7
Bowlder fescue
8
Hordeum jubatum
Wild barley
10
Agropyron cristatum
Crested wheatgrass
11
Agropyron repens
Quackgrass
12
Agropyron pauciflorum
Slender wheatgrass
13
Western wheatgrass
14
Kochia scoparia
Kochia
15
Golden annual sweet clover
16
Alysicarpus vaginalis
Alyce clover
17
Lespedeza sericea
Chinese lespedeza
18
Lespedeza stipulacea
Korean lespedeza
19
Crotalaria intermedia
20
21
Euphorbia corollata
Flowering spurge
22
Solanum carolinense
Horse nettle
23
Physalis heterophylla
Clammy groundcherry
24
Xanthium
Cocklebur

Fig. 20.

Fig. 21.

Seedlings of the white Dutch form produce no such reaction (13).

(d) Examination of seedlings.

Seeds of certain varieties of soybeans resemble each other so closely that identification by seed characters is impossible. In some of the black-seeded varieties such as Wilson and Laredo it is possible to grow young seedlings and by study of the cotyledons and first set of true leaves to distinguish between varieties (11).

Seedlings of European red clover have hairs appressed close to the stem whereas those of the American strains

have hairs that stand out prominently perpendicular to the stem (20).

Seeds of varieties and species within the genus Brassica are so similar that differentiation is possible only by seedling examination. Cabbage, cauliflower, broccoli, kale and turnip seedlings have definite leaf shapes that assist greatly in identification (24).

(e) Field tests. Many wheat, oat, barley and soybean varieties cannot be identified without growing them in the field to near maturity when the type of growth, degree of erectness, height, time of ripening and other agronomic characters can be observed.

(f) Use of pathological tests. The known reaction of wheat and oat varieties to physiologic races of rust and smut now makes it possible to either identify some of the new resistant varieties or to determine false labeling of lots claimed to be resistant. The relative resistance or susceptibility of varieties of cabbage, tomato, flax and watermelon to fusarial wilt organisms may be determined in the seedling stage. It is necessary to use either naturally or artificially-infested soil for such tests and to plant as checks, the seed of highly resistant and highly susceptible varieties for comparison with the unknown lot.

SPECIAL PROBLEMS IN SEED IDENTIFICATION

Certain crop seeds resemble weed seeds in the same genus or family so closely that differentiation is difficult. The resemblance between common weed seeds and those that are classed as noxious is also frequently close. This condition requires that whenever possible distinguishing features be described so that a reasonable degree of certainty can be achieved. The more important cases are as follows:

Barnyard (*Echinochloa crus-galli*) grass and Japanese millet (*Echinochloa frumentacea*);

Chess (*Bromus secalinus*) and other species of Bromus;

Dallis grass (*Paspalum dilatatum*) and other species of Paspalum;

Field bindweed (*Convolvulus arvensis*) and wild morning-glory (*Convolvulus sepium*);

Foxtails (*Chaetochloa spp.*) and millets (*Chaetochloa italica* and *Panicum miliaceum*);

Johnson grass (*Holcus halepensis*) and Sudan grass (*Holcus sorghum var. sudanensis*);

Meadow fescue (*Festuca elatior*) and ryegrass (*Lolium spp.*);

Quack grass (*Agropyron repens*) and wheat grasses (*Agropyron spp*);

Wild oats (*Avena fatua*) and false wild oats (*Avena sativa*);

Wild ground cherry (*Physalis spp.*) and horse nettle (*Solanum carolinense* and *S. elaeagnifolium*);

Wild mustard (*Brassica spp.*) and rape.

The more important features which assist in the differentiation of similar kinds of seed are given in tabular form in those cases when such an arrangement is considered helpful. In addition, the seed of each kind is illustrated in figs. 5 to 21. Arrangement of species and genera in each figure is by plant family.

DIFFERENTIATING FEATURES OF BARNYARD GRASS AND JAPANESE MILLET.

Name	Outer glumes	Size or shape of spikelet (and/or floret)	Color of lemma
Barnyard grass (*Echinochloa crus-galli*)	Outer glumes long, awn-pointed, margins usually with long bristles, sometimes short	Oval in outline, acute at apex, greatest width near center	Lemma green to gray, veins indistinct to noticeable
Japanese millet (*Echinochloa frumentacea*)	Outer glumes slightly awn-pointed with short teeth on the margins	Broad near base, short in relation to width, blunt at apex	Lemma gray, veins usually prominent

CHESS AND OTHER BROMUS SPECIES.

Name	Shape of fruit with glumes	Lemma	Naked fruit
Chess (*Bromus secalinus*)	Sturdy, somewhat cylindrical in outline, blunt at apex—folded	Short awned, wrinkled on back, adhering to fruit	Edges thick, usually folded in well toward the palea, sides straight
Japanese brome (*Bromus japonicus*)	Slightly broadened at apex. Slightly to prominently folded	Awned, smooth on back, wider than fruit	Edges thin, slightly curving or folded in toward palea
Downy brome (*Bromus tectorum*)	Long, slender curved backward on lemma side	Long awned, rough and hairy, adhering to fruit	Slender, curved, flattened or folded in slightly toward palea
Soft chess (*Bromus mollis*)	Broad at apex, flat, tapering toward base	Short awned if not broken, wrinkled on back	Usually flattened, seldom rolled inward
Smooth brome (*Bromus inermis*)	Usually broad at apex but tapering gradually toward base	Awnless	Usually flattened. Immature fruits sometimes rolled inward

DALLIS GRASS AND PASPALUM SPECIES

The fruits of Bahia grass (*Paspalum notatum*), *Paspalum laeve* and *Paspalum setaceum* are usually free from the outer glumes, the palea side is flattened and the lemma side is prominently humped. The fruits of dallis grass (*Paspalum dilatatum*) are usually covered with the outer glumes which are prominently hairy. The lemma side is rounded rather than humped.

FIELD BINDWEED (*Convolvulus arvensis*) AND WILD MORNING-GLORY (*Convolvulus sepium*)

The seeds of these two plants are rather distinctly different in size, field bindweed seeds being the smaller and more angular. In most cases the seeds of field bindweed have two flattened faces and a third face rounded and prominently humped. The surface of the seed coat is covered with tubercles which are readily seen under magnification. The seeds are broadest at the center, taper somewhat toward each end but more so toward the hilum which is almost on the narrow end. The seeds of wild morning-glory are irregular in shape, broad at one end and taper toward the hilum which is large and located silghtly above the point. The seeds are either angular or the edges are rounded and the seed coat surface is slightly to prominently roughened.

MEADOW FESCUE AND RYEGRASS (53)

Name	Rachilla	Palea	Lemma
Meadow fesque (*Festuca elatior*)	Rounded or oval in cross section; enlarged at apex; seldom appressed to palea, often bowed	Edges usually smooth, sometimes toothed	Awnless, usually buff colored, but occasionally light brown
Annual ryegrass. (*Lolium multiflorum*)	Flattened in cross section; sides mostly parallel; seldom enlarged at apex; usually appressed close to palea	Edges usually toothed, sometimes smooth	Awned unless broken; light brown to medium brown; seldom buff colored
Perennial ryegrass (*Lolium perenne*)	Similar to annual form but more uniformly appressed and not enlarged at apex	Similar to annual form	Awnless, usually light to dark brown, seldom buff colored

In size and shape the seeds of the ryegrasses and meadow fescue are similar. Immature or small seeds of meadow fescue often have slender, long and somewhat flattened rachillas.

JOHNSON GRASS AND SUDAN GRASS (18).

Name	Size and shape of:		Color of:		Pedicels
	Spikelet	Fruit	Spikelet	Fruit	
Johnson grass (*Holcus halepensis*)	Smaller than sudan, sharply pointed at apex, rounded at base	Greatest width above center, broad and blunt at apex, tapering toward embryo end	Mostly dark brown to black, occasionally buff	Amber, lustrous	Enlarged and cup-shaped at apex, sometimes hairy, frequently not
Sudan grass... (*Holcus sorghum var. sudanensis*)	Pointed at apex, tapering towards base	Greatest width at center, tapering toward each end	Usually buff, sometimes brown or or black	Dull, light to dark brown, greenish when immature	Ends jagged, not enlarged; sides usually hairy

FOXTAILS AND MILLETS.

Name	Shape of spikelet	Color	Lemma
Green foxtail.... (*Chaetochloa viridis*)	Oval, somewhat tapering toward apex; small	Green, mottled, brown, purple, almost black	Dull, sometimes ridged, frequently flattened above the callus
Yellow foxtail... (*Chaetochloa lutescens*)	Flat on palea side, humped on lemma; fruit large	Greenish, yellow or dark	Very rough on back with transverse ridges
Foxtail millet... (Hungarian, Siberian, Common, German) (*Chaetochloa italica*)	Broad and blunt as in German, somewhat tapering in others. Larger than seeds of green foxtail	Greenish, pale yellow, orange, purple, brown and almost black	Smooth, lustrous, veins faint to prominent as in German variety, flattened above callus
Proso or broom corn millet (*Panicum miliaceum*)	Broad, blunt, larger than any of the foxtail millets	Yellow, reddish, brown, purple, orange	Glossy, smooth, veins prominent

QUACK GRASS AND WHEAT GRASSES (17, 25)

Name	Rachilla	Palea	Lemma
Crested wheatgrass (*Agropyron cristatum*)	Usually short, thick and hollowed at end; stands out from palea	Edges usually with prominent teeth; margins folded toward each other	Keeled on back. Sharply awn-pointed, callus short and blunt
Quack grass.... (*Agropyron repens*)	Sides usually parallel; smooth or slightly rough; appressed close to palea	Tip rounded or slightly cup shaped, only partly exposed	Straight, awnless, awn-pointed or short-awned; prominent bulge above callus with no notch between
Slender wheatgrass (*Agropyron pauciflorum*)	Sides mostly parallel, sometimes broadened at apex, mostly appressed to palea; surface covered with soft hairs	Tip similar to quack grass but the palea is more exposed and flattened; distinct from lemma at apex	Offset from lemma, awnless, or awn-pointed; slight bulge above callus with no notch between; callus often hairy
Western wheatgrass (*Agropyron smithii*)	Broad at apex, narrow at base, not appressed close to palea; surface often slightly hairy	Tip with a cleft at apex; more open, broad and flat than in quack-grass	Straight, awnless or awn-pointed. Base not bulged but with a prominent notch between it and callus

WILD OATS (*Avena fatua*) AND

FALSE WILD OATS (*Avena sativa var.*) (26)

Externally the seeds of true and false wild oats are very similar. Both may be buff, brown or black; they have a twisted awn arising from the back of the lemma; the callus and rachilla are usually covered with hairs, and the base of the callus is large and open like a "sucker mouth." In size the false wild oats are usually somewhat larger than true wild oats but this difference is not uniform and is undependable as a means of differentiation. The best information to date is that false wild oats arise by mutation from cultivated varieties. They are, therefore, classed as crop seed, while wild oats are classed as a weed and in many seed laws are classed as noxious, hence differentiation is important.

The only reliable method of differentiation is to examine the naked grain. The naked grain of the false wild oat has a prominent scutellum above the embryo proper while in the true wild oat this region is not discernible. Figure 21, No. 2, illustrates the difference.

WILD GROUND CHERRY, HORSE NETTLE AND WHITE HORSE NETTLE (*Purple nightshade*)

In several of the northern states the seeds of horse nettle, *Solanum carolinense,* are classed as noxious, in some of the southern states the seeds of white horse nettle, *Solanum elaeagnifolium,* are noxious and in one of the western states the seeds of ground cherry, *Physalis subglabrata,* are classed as noxious. Seeds of all three species are given in figs. 20 and 21. Seeds of *Physalis subglabrata* and of several other species of wild ground cherry are slightly smaller than those of the common horse nettle and considerably smaller than white horse nettle seeds. With one known exception, wild ground cherry seeds are lemon yellow (one species has purple seeds), quite uniformly rounded in shape, and the surface of the seed coat is noticeably pitted as viewed under a magnifier. Horse nettle seeds are normally yellowish brown or dirty brown, irregular in shape, but more commonly narrower at one end than at the other, and the seed coat surface is usually smooth, seldom faintly pitted. Seeds of white horse nettle are nearly 1/8 inch in diameter, somewhat rounded or narrowed at one end, dark yellow or dirty brown in color and the seed coat surface relatively smooth.

WILD MUSTARDS AND RAPE

Seed of three common species of mustard are frequently classed as noxious in some midwestern states. These species are black mustard (*Brassica nigra*), Indian mustard (*Brassica juncea*) and wild mustard (*Brassica arvensis*). Seeds of Dwarf Essex and other varieties of rape at first glance appear similar to some or all of these mustards. Differences are given in tabular form as follows:

Name	Shape and size of seed	Color	Seed coat surface
Dwarf essex rape (*Brassica napus var.*)	Irregular but mostly spherical or flattened on one side. Mostly larger than the mustards	Mostly black, occasionally brown	Somewhat pitted, but not prominently
Black mustard.. (*Brassica nigra*)	Oval, smallest of three mustards	Brown and often covered with a whitish membrane	Prominently pitted
Indian mustard. (*Brassica juncea*)	Oval or sub-spherical	Brown	Prominently pitted
Wild mustard... (*Brassica arvensis*)	Spherical, small, fairly uniform in shape and size	Black when mature, brown when immature	Smooth or faintly pitted

GERMINATION TESTS

The primary purpose of a germination test by a seed laboratory is to determine the ability of seeds to grow and produce plants. It is necessary to have a standard by which seed viability may be measured. By some it is held that tests in soil under conditions as natural as possible should constitute the standard. Others have attempted to provide laboratory or greenhouse conditions intermediate between optimum and unfavorable conditions. The major difficulty in such approaches to the problem is that field conditions are too variable to allow the use of natural soil as a medium for germination. Drainage, fertility, physical condition, chemical reaction, amount of water and the fungous flora of soils are such variable factors that it would be impossible to provide a laboratory environment closely approaching natural field conditions.

The safest standard for germination tests would appear to be optimum conditions for each species or kind. This standard should measure the maximum viability. Field response may seldom be expected to equal that obtained in the laboratory, although many published records show that occasionally the percentage germination in the field of a given lot may equal or even exceed the percentage of normal seedlings obtained in the laboratory. The field germination of the larger seeds such as corn, beans, peas, pumpkin, squash, soybean, watermelon and the small grains approaches more nearly to the laboratory index than does that of small seeds such as clovers and grasses. If this proposed standard is employed then it should be possible to grade lots of seed of a given kind as to their relative ability to produce a crop in the field. There are specific cases, however, in which it is desirable to know the response of seeds to unfavorable conditions. For this purpose certain special tests have been provided. They will be described later under "cold tests" or "tests for diseases."

The sections on hard seeds, dormant seeds, pathological tests and interpretation of tests will assist in an understanding of the problems involved in determining the ability of seeds to produce plants.

HOW TO MAKE TESTS FOR GERMINATION

There are several basic requirements for seed germination that must be met if maximum viability is to be determined. They are:

(a) Adequate supply of water, (b) favorable temperature, (c) supply of oxygen, (d) favorable seedbed, (e) permeable seed coats and non-dormant embryos and (f) absence or control of highly parasitic organisms.

In addition to the above requirements some seeds require light, some an acid medium and others the addition of nitrates such as a dilute solution (0.2 percent) of potassium nitrate.

Accurate counting of seeds for the germination test is important. A vacuum pump to which a hose, valve and counter plate are attached serves well to count seeds ranging in size from redtop to wheat, oats and sorghum. The plate may have 100 or 200 holes at each of which a seed may be held by suction until placed on the germination medium. Larger seeds may be counted by means of counter boards as shown in figs. 22 and 23.

Materials used as a seedbed include blotters, towels, kimpack, sawdust, sand and soil. For small seeds of field, flower and vegetable crops, blotters are most commonly used. The seeds are placed either between one fold of blotters or on top of a blotter in a covered petri dish. Rolled paper towels, kimpack, sawdust, sand and soil are used for such large seeds as corn, peas, beans, pumpkin, squash and cereals.

Within recent years sand has received major attention as a medium for the germination of many kinds of seed. The Iowa State College Seed Laboratory has conducted extensive studies with sand as a medium for the germination of many kinds of seed with the result that sand has been adopted by the laboratory

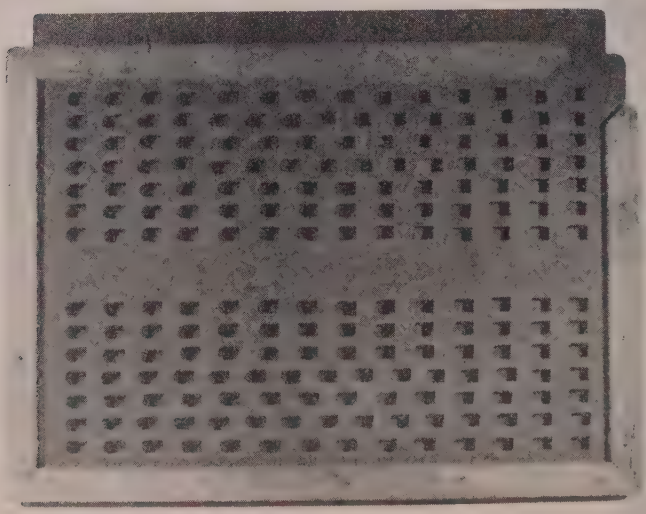

Fig. 22. A 200-hole counting board for corn.

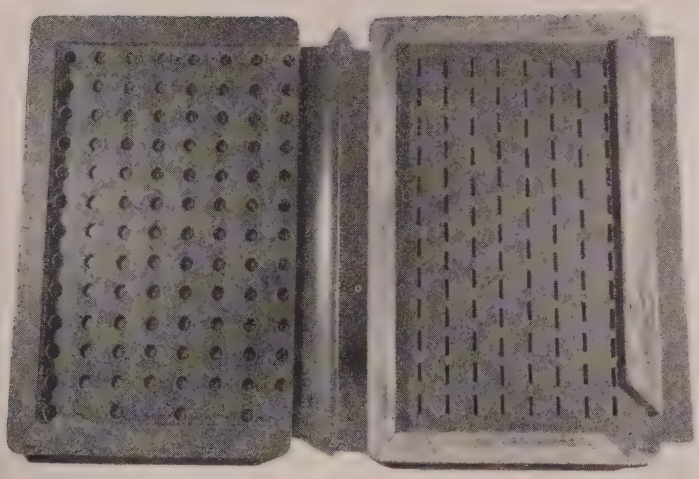

Fig. 23. Seed counting boards. Left—peas and soybeans. Right—cucumber and melons.

as a standard medium for such seeds as corn, soybean, garden bean and pea, pumpkin, squash, watermelon, cantaloupe, cucumber, horse bean, cowpea, barley, oats, wheat, rye, sorghum, vetch, field pea, bluegrass and reed canary grass. Builders' sand properly sterilized and placed in pans, flats, or benches serves as the medium for all large seeds. Organisms in the sand are controlled by subjection of the sand to radiated heat from either high pressure steam pipes or electric heating elements enclosed in a wooden container lined with galvanized iron. Two types of sterilizers are illustrated in figs. 24 and 25. The sand is saturated with water and then heated for not less than 15 hours at a temperature of about 70°C. Germination of seeds in sand benches is illustrated in fig. 26.

Seeds of bluegrass (31) and reed canary grass are tested on top of either screened river sand free from clay, or quartz sand (Flint Silica or Black Hawk No. 2) placed in covered petri dishes. The sand is heated in an oven over an electric plate for 6 hours and then moistened with distilled water to slightly above the saturation point. The same grade of quartz sand is also used to measure the emergence of seedlings of clovers, alfalfa, flax, lespedeza, onion, cabbage and other Brassica species. The sand is placed in copper boxes 4½″ x 9″ x 1½″ and the seeds are arranged between two layers with the top layer of sand about ½ inch thick. The sand is covered with a moist blotter to which water may be added as needed. When the majority of the

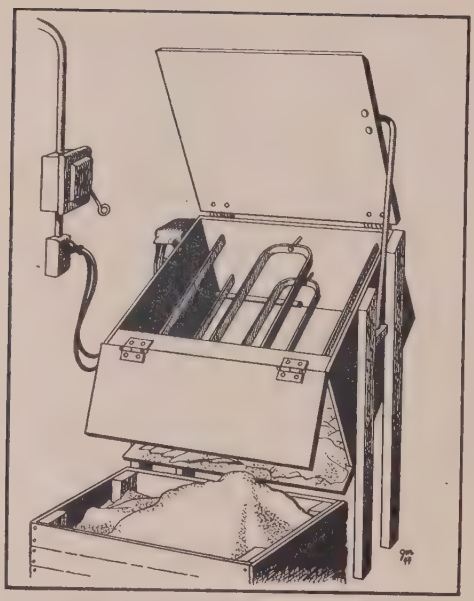

Fig. 24. Electric sand pasteurizer.

seedlings have emerged sufficiently to raise the blotter above the sand the blotter should be removed. At the end of the required period the seedlings may be pulled and examined for the presence of essential structures. Spinach seed may be planted on top of a layer of moist sand and covered only with a blotter which should be kept moist for the duration of the test.

The duration of the test, the temperature requirements, the number of seeds to plant, the substrata to use and other special requirements for several kinds of seed are given in table 4. *With few exceptions the data in table 4 are the same as given in the Rules for Seed Testing under the Federal Seed Act. The exceptions are based on the results of experiments conducted in the Iowa State College Seed Laboratory. Labeling of seeds for shipment in interstate commerce should be*

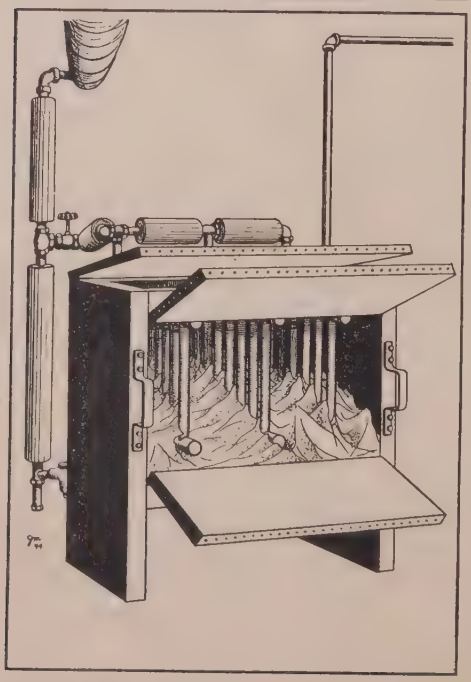

Fig. 25. Sand pasteurizer with steam coils.

Fig. 26. Sand bench for seed germination.

based on tests made according to federal rules in all cases.

DETERMINATION OF HARD SEEDS

Impermeable or hard seeds occur commonly in samples of clovers, alfalfa, lespedeza, peas, beans, soybeans, vetch, morning-glory, okra and asparagus seeds. The presence of such seeds in germination tests may be readily detected in blotters or towels. These seeds are firm or hard, unswollen, usually bright, fresh in appearance and relatively free from molds. They are distinct from dead seeds, which are often moldy, mushy and decayed. All hard seeds should be counted and recorded in each test. In some cases these seeds, which vary in the degree of impermeability, may be slightly swollen at the end of a test and if left longer will germinate. Experience with these seeds will sharpen one's judgment in recognizing them and distinguishing them from dead seeds. They should be included with the hard seed count. It is possible to determine the number of unswollen or hard seeds of the large seeds such as peas and beans in sand tests. They are large enough to be observed when the sand is stirred after the seedlings are removed.

The causes for impermeable seeds are not completely understood. Many theories have been advanced, but from the prac-

TABLE 4. TEMPERATURE, SUBSTRATA AND OTHER SPECIAL REQUIREMENTS FOR LABORATORY GERMINATION OF SEEDS.

Name of seed	Substrata*	Temperature °C.	Days after planting: First count	Days after planting: Second count	Special methods
1. Field crops					
Alfalfa	B, Sa	20	3	7	In sand make one count
Barley	B, S	20	3	7 or 8	Prechill fresh seed 10°C. 5 days
Beet	B	20-30	3	14	Soak in water 2 hrs., wash in running water, then plant
Bentgrass					
Creeping (seaside)	P	20-30	7	28	Potassium nitrate[1]; light
Highland	P	20-30	7	28	Potassium nitrate[1]; light
Others	P	20-30	7	21	Potassium nitrate[1]; light
Bluegrass					
Canada	PS	20-30	10	28	Potassium nitrate; light
Kentucky	PS	20-30	10	28	Light; chill fresh seed. 0.1% KNO_3
Rough	PS	20-30	10	21	Light; chill fresh seed. 0.1% KNO_3
Bromegrass	B	20-30	6	14	Light for fresh seed.
Buckwheat	B	20-30	3	6	
Clovers					
(a) Alyce	B, Sa	30	7	21	
(b) Alsike, crimson	B, Sa	20	....	....	
Persian, red	B, Sa	20	....	....	Sweet clover, crimson and red clover in sand one count
Sweet, strawberry	B, Sa	20	3	7	
(c) Sour, suckling	B	20	3	14	
(d) White	B	20	3	10	
Dogtail	P	20-30	10	21	Light; chill fresh seed 3 days 10°C.
Cowpeas	S	20-30	3	10	
Corn, all kinds	S	20-30 or 30	....	7 or 6	
Crotalaria	B, Sa	20-30	3	10	
Fescue					
Hair	P	10-25	10	28	Potassium nitrate
Meadow	P	20-30	5	14	
Others	P	15-25	7	21	
Flax	B	20-30	3	7	
Grass					
Bermuda	P	20-35	7	21	Potassium nitrate; light
Carpet	P	20-35	10	21	Potassium nitrate; light 8 hours daily
Dallis	P	20-35	7	21	Potassium nitrate; light
Johnson	P	25-40	5	18	Sulfuric acid 30 min.
Orchard	P	20-30	7	18	Light; excess of water
Reed canary	PS	20-30	5	21	Light; fresh seed potassium nitrate
Sudan	B	20-30	3	10	Use sand for treated seed
Hemp	B, Sa	20-30	3	7	
Lespedeza					
Chinese	B	20-35	7	28	
Common and Korean	B	20-35	7	14	
Lupine	B, S	20	7	21	
Medick, black	B	20	3	7	
Millet					
Broom corn (proso)	B	20-30	3	7	
Brown top	B	20-30	4	14	Fresh seed often responds to chilling
Cattail (Pearl)	B	20-30	3	7	
Foxtail	B	20-30	4	10	Fresh seed often responds to chilling
Japanese	B	20-30	4	10	
Oatgrass, tall	P	20-30	6	14	Light
Oats	B, S	20	4	10	Chill dormant seed 5 days 10°C.
Peas (field)	S	20	....	10	
Rape	B	20-30	3	7	10 days for bird rape with light and KNO_3
Red top	P	20-30	5	10	Light
Rye	S	20	....	10	
Ryegrass	P	20-30	5	14	Light; chill fresh seed 5 days 10°C.
Sainfoin	B	20-30	4	10	
Sorghum	B, S	20-30	....	....	Use sand for treated seed
		or 30	3	10	Chill fresh seed of sweet varieties 5 days 10°C.
Soybean	S	20-30 or 30	5	8 or 6	
Timothy	P	20-30	5	10	Light; chill fresh seed 3 days 10°C.

TABLE 4—(Continued)

Name of seed	Substrata*	Temperature °C.	Days after planting		Special methods
			First count	Second count	
Trefoil, birdsfoot	B	20	3	7	
Vetch					
(a) Common, Hungarian and purple	B, S	20	3	10	
(b) Hairy, narrow-leaf and wooly pod	B, S	20	4	14	
Wheat					
Common	B, S	20	3	7 or 8	Chill fresh seed 5 days 10°C.
Durum	B, S	20	3	10	Chill fresh seed 5 days 10°C.
Wheat grass					
Crested	P	20-30	5	14	Light; fresh seed 15°C.
Slender	B	20-30	5	14	
Western	P	20-30	10	35	Potassium nitrate
2. Vegetables and herbs					
Asparagus	B, S	20-30	7	21	
Beans					
Garden	S	20-30 or 30	5	10	
Horse	S	20	5	10	
Lima	S	20-30 or 30	5	10	
Beet, Swiss chard and mangel	B	20-30	3	14	
Brassica oleracea	B	20-30	3	10	
Cabbage, collards, kale, brussels sprouts, cauliflower, broccoli and kohlrabi					Fresh seed often responds to potassium nitrate and chilling
Carrot	P, B	20-30	6	28	
Celeriac and celery	P	20-30	10	21	Light
Chicory	P	20-30	5	14	Light; potassium nitrate
Citron	B, S	20-30	7	14	
Corn salad	P	20	7	28	Fresh seed at 10° or 15°C.
Cress	P	20	4	10	
Cucumber and cantaloupe	S	20-30 or 30	....	10 or 7	
Dandelion	P	20	7	21	
Dill	B	20-30	7	21	
Egg plant	P	20-30	7	14	
Endive	P	20	5	14	Light; potassium nitrate
Lettuce	P	20	3	5	Light; 2 hrs. daily; chill fresh seed 3 days 10°C.
Mustard, India	P	20-30	3	7	Light; chill fresh seed with nitrate
Okra	B	....	4	21	
Onion and leek	B, S	20	5	10	Only one count in sand
Parsley	P	20-30	11	28	
Parsnip	B	20-30	6	28	
Peas	S	20	....	8	
Pepper	B	20-30	6	14	
Pumpkin	S	20-30 or 30	4	7	
Radish	B	20	3	6	
Rhubarb	Top soil	20-30	7	21	Light
Rutabaga	B	20-30	3	7	
Salsify	B, S	20	5	10	Chill fresh seed 3 days 10°C.
Sorrel	Top soil	20-30	3	14	Light
Spinach					
Common	Top sand	10	7	21	Cover with blotter
New Zealand	Top soil	20-30	5	28	Not too wet
Squash	S	20-30 or 30	4	7	
Tomato					
Common	B	20-30	5	14	
Husk (ground cherry)	P	20-30	7	28	Light
Turnip	B	20-30	3	7	
Watermelon	S	20-30 or 30	....	14 or 10	Many samples completed (90 to 95% germination) in 7 to 10 days at 30°C.

* B=Blotters (folded); P=Petri dish with 2 thicknesses of blotters; PS=Petri dish with quartz sand; S=Builder's sand in benches, pans or flats; Sa=quartz sand in small pans or paraffin paper boxes.

† 5° C.=41°F. 10°C.=50°F. 15°C.=59°F. 20°C.=68°F. 25°C.=77°F 30°C.=86°F. 35°C.=95°F.

[1] Potassium nitrate (KNO_3) is prepared by dissolving 2 grams in 1 liter (1000 ml.) of water which makes 0.2 percent solution. For Kentucky bluegrass use 0.1 percent.

It is recommended that 400 seeds be used for each germination test whenever possible.

tical standpoint the important thing to know is that an area in the layers of the seed coat is impervious to water and without water germination is impossible.

TREATMENT OF DORMANT SEEDS

Seeds of many grasses, some of the cereals, certain varieties of lettuce, crucifers and many kinds of trees and shrubs are frequently dormant when harvested. The most common causes (7) of dormancy are:

a. Immature and undeveloped embryos.
b. Seed coats impervious to entrance of water.
c. Covering of embryo, seed coats, or pericarp prevents adequate exchange of carbon dioxide and oxygen for germination.
d. Seed or fruit covering too tough to permit expansion of embryo and emergence of radicle.

In germination tests it is possible, with some experience, to differentiate dormant from dead seeds. Dormant seeds are usually free from mold, fairly firm, bright and fresh in appearance. They are not mushy and decayed but they can be crushed with tweezers or between the fingers whereas hard seeds cannot.

Freshly harvested seeds of wheat, barley, oats, bluegrass, orchard grass, ryegrass, meadow fescue, bentgrass, sorghum, sudan grass and millet are often dormant. Seeds of the new oat varieties—Tama, Boone and Vicland—are usually dormant when harvested and dormancy often persists until the spring months.

There are six common methods that may be employed to break seed dormancy and bring about fairly prompt germination. Drying of the seeds either naturally or with artificial heat often breaks the dormancy of grains and grasses. Exposure for 24 hours in an oven at 105° to 108°F. is usually sufficient time. A second method most successfully used with cereals and grasses (50) is to place the seeds in a moist condition at a temperature of 40° to 50°F. for 3 to 5 days after which they may be transferred to the temperature that is favorable for the germination of non-dormant seeds. Dormant seeds of trees and shrubs are commonly stratified by storage between layers of moist peat moss for several months with temperatures slightly above freezing. The other three methods consist of treatment with sulfuric acid to remove outer seed or fruit coverings, addition of 0.2 percent potassium nitrate solution to the seeds in place of water, and exposure to light. Combinations of light and nitrate, light and low temperature or other combinations are also employed. Low temperature and light are required for dormant lettuce seed, light and nitrate for Canada bluegrass,

bent grasses and some crucifers, and low temperature and nitrate are frequently beneficial for freshly harvested Kentucky bluegrass seed.

PATHOLOGICAL TESTS

DETECTION OF SEED-BORNE ORGANISMS

Within recent years a few seed analysts (8, 27, 30, 32) have placed considerable emphasis on the need for including in the service of seed testing the determination of organisms carried by seed lots, especially those of a pathogenic nature capable of inflicting serious losses on the subsequent crop. It has been recognized, for example, that many seed lots which possess high purity and high germination carry organisms that cause serious crop losses. Other organisms may be primarily responsible for reduced germination. Recognition of the relation of disease-producing organisms to the practice of seed testing has resulted in the application of certain pathological techniques to seed laboratory procedures.

Three major techniques have been employed (a) development of practical methods for determining the presence of seed-borne organisms, (b) identification of the specific organisms and (c) measurement or interpretation of the significance of the organisms found.

Determination of seed-borne organisms in a modern seed laboratory is accomplished primarily by four methods (30). The first involves washing of seeds in sterile water either by shaking or centrifuging, after which microscopic examination of the washings is made to determine what fungous spores are present (12). This method is simple and rapid and although it does not reveal the percentage of seeds infected it does give some indication of the prevalence of organisms on the seed. It is even possible to measure the spore load by the use of a haemocytometer (41). This method is especially applicable to the detection of smut spores carried by seeds of cereal grains, millet and sorghum.

A second method consists in either plating on sterile agar seeds that may or may not be surface disinfected or in planting seeds properly spaced on top of moist blotters in large petri dishes. If such tests are conducted at the proper temperature it is usually possible to determine if certain specific organisms are present, the approximate percentage of infected seeds and an estimate of the probable injury to germination. This method is especially applicable to seed corn infected with dry rot fungi, to wheat, barley, oats and sorghum infected with seedling blight organisms, to peas infected with pea blight organisms and to soybean and garden bean seeds infected with anthracnose fungi. In some cases the wilt producing organisms on seeds of cabbage,

flax, tomato and watermelon can be detected by this method.

A third method involves planting seeds in autoclaved soil or sand, either in a greenhouse or laboratory, maintaining the proper temperature and humidity, and noting such symptoms as wilt and seedling blight after the seedlings have emerged. The presence of organisms that cause seedling blights of corn, wheat, barley, oats, sorghum, beans and cabbage, and of those that cause fusarial wilts of cabbage, flax, tomato and watermelon can be detected with reasonable accuracy by this method.

A fourth method has to do with the detection of virus diseases either in seeds or vegetative, propagative organs of plants. This method requires planting of seeds, bulbs or tubers in a greenhouse in which temperature and light are adequately controlled to effect an expression of virus disease symptoms. Mosaic of potatoes, yellow dwarf of onions and mosaic of beans are some of the diseases whose symptoms can be detected by this method.

A partial list of the organisms or viruses whose presence in seed stocks may be detected in a seed laboratory is given in table 5. The identification of new or unfamiliar organisms requires the assistance of a mycologist and the employment of well known laboratory techniques which may cause the appearance of fruiting structures that aid in the identification of organisms. Measurement or evaluation of probable injury that organisms may cause requires experience and familiarity with the conditions that determine the expression of disease. Field experiments comparing the data from laboratory studies with field response have been employed with some success (32) in evaluating the significance of seed-borne organisms and have thus aided the seed analyst in subsequent observations. In general, when smut spores are present on seeds or when seedling blight fungi are carried by seeds it may be expected that the particular diseases resulting from such infection will appear in fields sown with the seed and the damage caused will depend to a considerable extent on environmental conditions.

REACTION OF SEEDS TO SOIL-BORNE ORGANISMS

Within the past few years important studies have been made of the effect of such soil-borne organisms as Pythium species on field stand (5, 6, 19). An effort has been made in the Iowa State College Seed Laboratory (40) to apply the results of these investigations to seed laboratory practice. Specifically, many hundreds of samples of hybrid seed corn have been planted in a mixture containing equal parts of sand and unsterilized soil known to contain pythiaceous fungi. These samples are held for 7 days in this moist soil and sand mixture at a constant temperature of 50°F. after which a transfer is made to a room with a temperature of 75°F. After 3 or 4 days at the latter

temperature the samples are examined and counts made for emerged, normal seedlings. Marked differences have been found in the response of seed lots to injury caused by soil fungi. Differences have not been confined to lots of different inheritance. Tatum and Zuber (49) and Rice (42) have shown that the treatment which corn receives during its processing for seed has a marked effect on its germination in unsterilized soil at 10°C. From the standpoint of a seed laboratory it is most important to find out which lots each year are most likely to be injured in the field, and a testing service is offered for that purpose. A most important requirement is to start each test with the same amount of moisture, the determination of which can be greatly aided by the use of a soil tensiometer (37).

In 1942 data were kept on 177 lots of hybrid corn. The mean percentage germination in sterile sand at daily temperatures ranging from 68°F. to 86°F. was 92. The same samples subjected to the cold test in natural soil as previously described gave a mean percentage germination of 67.5. Seeds of soybeans, sorghum, hemp, barley, flax, peas, spinach, onion, beet and sweet corn (23) respond in a similar manner although the number of days of exposure to 50°F. for the cold test is not necessarily the same for each crop.

USE OF SEED DISINFECTANTS AND PROTECTANTS

The development and use of chemical dusts for the pretreatment of seeds have been rapid and extensive within the past decade. Not only have new synthetic compounds been developed that have proved effective for the control of seed-borne organisms but many have proved effective protectants against the attack of soil-borne organisms. In a number of cases these compounds possess the properties of both disinfection and protection. Use of seed disinfectants and protectants in seed testing procedures has also received considerable attention (9, 27, 28, 32).

Treated seeds of barley, oats and wheat if infected with the scab fungus (*Gibberella saubinetii*) and planted on top of moist blotters, have been shown to exhibit a growth of pink mycelium (mold) around each infected seed. The same seed if treated with a volatile mercury compound such as Ceresan or New Improved Ceresan will be relatively free from fungous growth. If such seed is planted in a mixture of autoclaved soil and sand, the seedlings produced by untreated seed will usually show marked symptoms of seedling blight, those from treated seed will remain relatively free from blight and furthermore the treated seed will produce from 10 to 50 percent more seedlings.

Results similar to those described for small grains may be obtained with seed corn infected with the dry rot fungi *Diplodia*

TABLE 5. LIST OF SEED-BORNE ORGANISMS DETECTABLE IN OR ON SEEDS.

Crop	Common name of disease	Causal organism	How carried by seed	Signs or symptoms on seeds, seedlings or older plants
1. FIELD CROPS				
Barley	Covered smut	*Ustilago hordei*	Smut balls; spores on seed	Smut balls; spores in washing
	Loose smut	*Ustilago nuda*	Inside seed	Smutted heads on greenhouse plants
	Scab	*Gibberella saubinetii*	Fruiting bodies, spores and mycelium	Pink mold on seeds on blotters; blighted seedlings in soil
	Seedling blight	*Helminthosporium sativum*	Mycelium and spores	Brownish roots and stems of seedlings on blotters
	Stripe	*Helminthosporium gramineum*	Mycelium	Stripes of yellow and brown on greenhouse seedlings—seed germinated at 12°C
Beet	Mosaic	Virus	Virus in young roots	Mottling of young leaves of greenhouse plants
Bentgrass	Ergot	*Claviceps sp.*	Black ergot bodies	Readily seen with normal seeds
Bluegrass	Ergot	Same as for bentgrass		
Bromegrass	Scab	*Fusarium sp.*	Spores and mycelium on seeds	Similar to barley
Carpetgrass	Ergot	Same as for bentgrass		
Corn	Dry rot	*Diplodia zeae*	Fruiting bodies and mycelium	White mold on germinating seeds; seedling blight
	Dry rot	*Gibberella saubinetii*	Spores and mycelium	Pink and white mold on seeds and seedlings; seedling blight
	Dry rot	*Nigrospora oryzae*	Spores and mycelium	Large black spores on kernels; rotted seeds at low temperatures
	Black bundle	*Cephalosporium acrimonium*	Spores and mycelium	White to salmon-colored fungus on seeds
Cotton	Anthracnose	*Gloeosporium gossypii*	Spores and mycelium	Pink mold on seeds; blighted seedlings
Dallis grass	Ergot	*Claviceps sp.*	Black growth in and on seeds	
Millet	Smut	*Ustilago crameri*	Spores	Spores in washings
	Downy mildew	*Sclerospora graminicola*	Spores	Large oospores in washings
Oats	Smut	*Ustilago avenae*	Spores	Spores in washings
	Scab	Same as for barley		
	Seedling blight	*Helminthosporium spp.*	Same as for barley	
Redtop	Ergot	Same as for bentgrass		
Rice	Smut	*Tilletia horrida*	Smut balls; spores	Spores in washings
Rye	Ergot	*Claviceps purpurea*	Same as for bentgrass	
	Scab	Same as for barley		
	Seedling blight	Same as for barley		
Ryegrass	Ergot	Same as for bentgrass		
	Scab	Same as for barley		

TABLE 5– (Continued)

Crop	Common name of disease	Causal organism	How carried by seed	Signs or symptoms on seeds, seedlings or older plants
Sorghum	Blight	*Helminthosporium sp*	Mycelium	Spores on blighted seedlings
	Blight	*Fusarium spp.*	Mycelium	White mold on seeds and seedlings
	Smut—covered	*Sphacelotheca sorghi*	Spores and smut balls	Spores in washings
	Smut—loose	*Sphacelotheca cruenta*	Spores and smut balls	Spores in washings
Soybean	Anthracnose	*Colletotrichum glycine*	Mycelium	Spots on cotyledons; blighted seedlings
Timothy	Scab	*Fusarium sp.*	Same as for barley	
	Seedling blight	*Helminthosporium sp.*	Mycelium	Spores on blighted seedlings
Wheat	Stinking smut	*Tilletia sp.*	Smut balls and spores	Odor of dead fish—spores in washings; smut balls
	Smut (flag)	*Urocystis tritici*	Spores	Spores in washings
	Smut (loose)	*Ustilago tritici*	Mycelium in seed	Same as for loose smut of barley
	Scab	Same as for barley		
2. VEGETABLE CROPS	Seedling blight	Same as for barley		
Beans	Anthracnose	*Colletotrichum lindemuthianum*	Mycelium	Spots on seeds; lesions on young seedlings
	Blight	*Phytomonas spp.*	Bacteria on seeds	Spots on seeds; wilting of seedlings; cankers on stems
	Mosaic	Virus	Virus in seeds	Mottling of leaves of greenhouse plants
Cabbage and other Brassicas	Black-leg	*Phoma lingam*	Spores and mycelium	White mold on seed; canker at base of young stem
	Black-rot	*Bacterium compestre*	Bacteria	Blackened areas on cotyledons and margins of young leaves
	Blight	*Alternaria brassicae*	Mycelium beneath seed coat	Black mycelium on seeds in germinator
	Yellows	*Fusarium conglutinans*	Mycelium and spores	White mold on seeds; wilted, yellowed or malformed leaves
Cucurbits	Anthracnose	*Colletotrichum lagenarium*	Spores and mycelium	Whitish mold on germinating seeds
Egg plant	Blight	*Phomopsis vexans*	Spores and mycelium	Whitish mold on seeds; lesions on seedlings
Onion	Yellow dwarf	Virus	Virus in bulbs	Yellowish plants in greenhouse flats
	Blight	*Mycosphaerella pinodes*	Spores and mycelium	Dense mycelium on seeds; lesions on seedlings
Potato	Black scurf	*Corticium vagum*	Sclerotia on tubers	Readily observed on tubers
	Scab	*Actinomyces scabies*	Bacteria in lesions on tubers	Small to large corky areas on tubers
	Mosaics	Virus	Virus in tubers	Mottling on greenhouse plants
	Spindle tuber	Virus	Virus in tubers	Spindly symptoms on greenhouse plants
Tomato	Blight	*Macrosporium spp.*	Spores and mycelium	Spots on leaves and stems of seedlings
	Wilt	*Fusarium lycopersici*	Spores and mycelium	Wilted seedlings at high temperature
Watermelon	Anthracnose	Same as cucurbit anthracnose		
	Wilt	*Fusarium niveum*	Mycelium	Wilted seedlings at high temperature

zeae, Nigrospora oryzae (36) and *Gibberella saubinetii,* and with sorghum seeds infected with species of *Helminthosporium* or *Fusarium.*

A further value of seed treatment is the control it affords of saprophytic organisms such as species of *Rhizopus, Penicillium, Aspergillus, Mucor* and *Alternaria* that often overrun samples of seed whose vitality is on the decline. The growth of such molds makes a reliable interpretation of seedlings quite difficult and is one of the marked causes of wide differences in tests reported by different laboratories. Seedlings from treated seed are more easily evaluated (38).

The protective value of seed treatment compounds may be determined in seed laboratory tests by planting treated and untreated seed in randomized lots using the same technique as described for cold tests on page 48. The data obtained from the 177 lots of hybrid seed corn using treated seed gave a mean percentage germination of 85.3. This figure is 17.8 percent above the untreated and only 6.7 percent lower than that obtained in sterile sand at very favorable temperatures for corn germination. Tests with 12 lots of soybean seed grown in 1943 using the cold test method with treated and untreated seed gave 58 percent normal seedlings from the untreated and 73.8 percent from the treated (40). Seeds of peas, sorghum, flax, hemp, spinach, onions, beets and sweet corn are likewise protected from the attack of soil fungi by application of certain compounds. A list of some of the more effective commercial dusts is given in table 6.

There can be no doubt but that the seeds of many field and vegetable crops may be greatly protected during critical germinating periods by the use of proper protectants. In general, when the soil moisture is high and the temperature favorable (50° to 60°C.) for infection by pythiaceous fungi then the greatest benefit from treatment of even good seed may be expected. Figures 27, 28 and 29 illustrate the beneficial effects of seed protectants to seeds of corn, peas and spinach when planted in Pythium-infested soil at a low temperature for the early part of the germination period.

HOW TO TREAT SEEDS IN A SEED LABORATORY

The purpose of treating seeds in a seed laboratory may be to (a) eliminate surface organisms so that the seeds may be plated in sterile agar to determine internal parasites, (b) control organisms that interfere with germination or the evaluation of sprouts, (c) evaluate seed disinfectants to compare with subsequent field performance and (d) control disease organisms on small seed lots for field planting. Treatments are either liquid or dry.

Fig. 27. Cold test of corn seed. Left—untreated. Right—treated with Spergon.

Fig. 28. Cold test of peas. Left—untreated. Right—treated with Arasan.

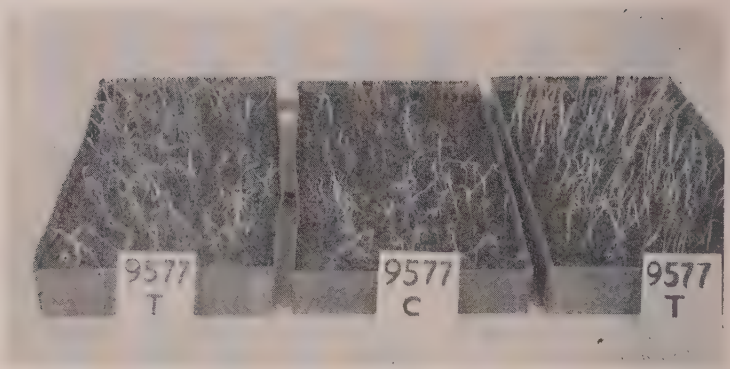

Fig. 29. Cold test of spinach seed. Left—treated with Arasan. Center—untreated. Right—treated with zinc oxide.

LIQUID SEED DISINFECTANTS

Liquid treatments generally are used to achieve either the first or fourth purpose mentioned in the preceding paragraph. The more common ones, their preparation and use are as follows:

(a) Hydrogen peroxide—30 percent concentration. One part of this solution to 9 parts distilled water is a satisfactory dilution. Seeds may be soaked 5 or more minutes, dried in a sterile dish and then plated in sterile agar or planted on blotters.

(b) Ch'orox, 5.25 percent strength. Dilution and use of this solution should be the same as for hydrogen peroxide.

(c) Calcium hypochlorite. Dissolve 10 grams of commercial chloride of lime in 140 milliliters of water, allow to settle, decant the liquid and use as a disinfectant without dilution. Seeds so treated may be transferred directly to sterile agar or to blotters. The duration of treatment varies with the kind of seed (52).

(d) Bichloride of mercury. This chemical is deadly poison and should be used with great care. The common strength is 1 gram in 1000 milliliters of water, except for tomato seed for which the strength is 1 to 3,000. Seeds may be soaked 5 to 15 minutes in this solution, then rinsed three times in sterile water and plated in sterile agar or planted in blotters, sand or soil.

(e) Formaldehyde (37 percent). A common dilution is 1 part to 240 parts of water. Seeds may be soaked for 5 to 20 minutes, then dried and planted in blotters, sand or soil.

(f) Hot water. Treatment of wheat for 10 to 12 minutes in hot water at a temperature of 54°C. is effective in the

control of loose smut. Pre-treatment in cold water, then 1 minute at 45°C. is beneficial. Loose smut of barley is also controllable with hot water. Treatment of cabbage seed to kill the black leg and black rot organisms is best accomplished with hot water. Soaking the seed 20 minutes in water at 50°C. is effective but the seed must be cooled at once and dried. It is also necessary to use a water bath with almost absolute temperature control. Tomato seed may be treated by soaking in water for 30 minutes at 50°C. Careful control of the temperature is necessary.

DUST TREATMENTS

A list of dust treatments used for seeds is given in table 6. Application of these dusts to seeds can be made by (a) shaking the seeds with the required amount of dust in a bottle (b) rotating the bottles on a laboratory machine illustrated in fig. 30 or (c) by use of the seed mixer illustrated in fig. 2.

The safest procedure is to calculate and weigh the amount of

TABLE 6. LIST OF SEED DISINFECTANTS AND PROTECTANTS USED IN SEED TESTING AND FIELD PLANTING.

Name of material	Crop	Dosage	Purpose
Arasan	Bean	⅓ tsp. per 2 oz. seed	Seed protectant
	Beet	⅓ tsp. per 2 oz. seed	Seed protectant
	Corn (field)	1½ oz. per bu. seed	Seed protectant
	Corn (sweet)	⅓ tsp. per 2 oz. seed	Seed protectant
	Pea	⅔ tsp. per 4 oz. seed 1½ oz. per bu.	Seed protectant
	Onion	⅔ tsp. per 4 oz. seed	Seed protectant
	Spinach	⅔ tsp. per 4 oz. seed	Seed protectant
	Sorghum	1 oz. per bu.	
Barabak C	Corn	1½ oz. per bu.	Seed protectant and fungicide for dry rot fungi
Ceresan	Sorghum	2 oz. per bu.	Smut control and seed protectant
Ceresan (New Improved)	Barley	½ oz. per bu.	Control of covered smut, stripe, seedling blight, damping off
	Beets	5 oz. per 100 lb.	Seed protectant
	Flax	½ oz. per bu.	Seed protectant
	Millet	½ oz. per bu.	Smut control
	Oats	½ oz. per bu.	Control smut, seedling blight, damping off
	Rye	½ oz. per bu.	Control of stinking and stem smuts, seedling blights
	Sorghum	½ oz. per bu.	Control kernel smuts, seedling blights
	Wheat	½ oz. per bu.	Control bunt, flag smut, seedling blights
Copper carbonate	Barley (hulless)	2 oz. per bu.	Covered smut and stripe
	Millet	2 oz. per bu.	Kernel smuts
	Oats (hulless)	2 oz. per bu.	Smuts
	Sorghum	2 oz. per bu.	Kernel smuts
	Wheat	2 oz. per bu.	Bunt, flag smut
Merko	Corn	1½ oz. per bu.	Control dry rot fungi seed protectant
Semesan Jr.	Corn	1½ oz. per bu.	Control dry rot fungi seed protectant
Spergon	Corn	2 oz. per bu.	Seed protectant
	Lima beans	3 oz. per bu.	Seed protectant
	Peas	2 oz. per bu.	Seed protectant
Zinc oxide	Spinach	2% by weight	Seed protectant

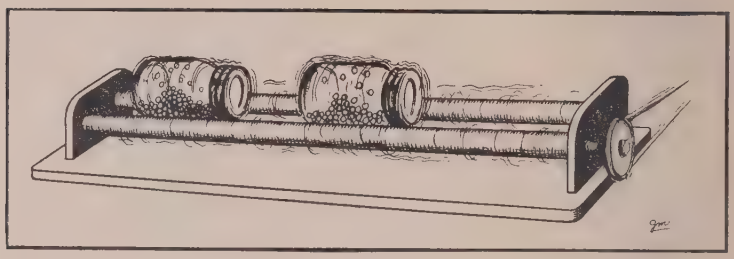

Fig. 30. Seed treater.

dust required and add this to the seed. For corn, wheat, rye, sorghum, beans, peas and melons it is reasonably safe to apply an excess of dust then screen off the excess, but for careful laboratory testing the more accurate weighing method should be used.

INTERPRETATION OF TESTS

One of the most difficult yet important tasks is to interpret the results of a test. This involves consideration of the purity and weed seed content, the viability, the hard seed content, the disease organisms carried by the seed and its resistance or susceptibility to organisms in the soil.

Other factors being equal it is obvious that seed lots with a relatively high purity are superior to others. The character of the impurities and the relative price, however, must receive consideration. If the impurities are other crop seeds they may even increase the seeding value but if they are weed seeds they reduce the value. The presence of noxious weed seeds may even disqualify the seed for sale, depending upon the requirements of state and federal seed laws. In Iowa and in many other states it is unlawful to sell seed containing seeds of primary noxious weeds. The use of seed lots containing seeds of secondary noxious weeds should be determined by the amount present and the availability of seed free from such weed seeds. The possibility of removing the weed seeds by recleaning must also be considered.

In viability tests it is necessary to consider the rate at which germination proceeds and the relative vigor of the sprouted seeds. In general, the older the seed the less vigorous the seedlings, thus indicating declining vitality. Abundant growth of saprophytic fungi also indicates declining vitality. In seed laboratory practice it is necessary to define germination and to classify seedlings into two major groups, normal and abnormal. Seed germination is defined as the emergence and development from the seed embryo of those essential structures which, for the

kind of plant in question, are indicative of the ability to produce a normal plant under favorable conditions.

In general, the essential structures as shown in fig. 31 are the primary root (radicle) and primary stem (plumule) each showing evidence of continuing growth under favorable conditions. There are examples among plants in which absence of the primary root does not prevent continued growth of a well anchored seedling. Corn seedlings frequently fail to produce a primary root, but the stem emerges normally, secondary roots are formed and the seedling develops into a plant equal in growth to those with a primary root at the time of germination. A similar condition often exists with onion, bean and melon seedlings. On the other hand, absence of a primary root in radish, carrot, beet and turnip seedlings would be serious. Soybean and garden bean seedlings frequently fail to develop the primary stem which normally forms between the two cotyledons that emerge from the soil. Absence of the young stem classifies the seedling as a bald head or abnormal seedling which, even though it may later produce a stem from a secondary bud, is inferior to a normal seedling. Many bald head plants are nearly equal to normal seedlings in their production of fruit and some allowance should probably be made for them in the report of germination. Investigation of their value is being continued but present rules require that they be classed as abnormal. Other types of abnormal seedlings are those with (a) a blunt undeveloped primary root, (b) cotyledons broken from the hypocotyl, (c) a watery (glassy) hypocotyl, (d) a split radicle or hypocotyl and (e) a rotted hypocotyl or plumule. Figures 31 to 44 illustrate normal and abnormal seedlings of several kinds of plants.

Viability is expressed in percentage germination which means the number of seeds out of each hundred that produce normal seedlings. Seeding rates frequently need to be adjusted according to the vitality of the seed. In addition, allowance should be made for the impermeable (hard) seeds (15, 51) that occur among members of leguminous and other plants. Hard seeds in alfalfa, lespedeza, soybean, garden bean, garden pea and okra usually germinate soon enough after planting to be equal to readily germinable seeds. In red, alsike, white Dutch and sweet clover from one-third to one-half the hard seeds may be considered as germinable the year they are planted. Fall or winter planting of clovers in some parts of the United States may result in a higher percentage germination of hard seeds.

APPLICATION OF SEED ANALYSES TO RATES OF SEEDING

The final seeding value of a given seed lot is determined in part by a combination of the purity and germination which is

best expressed by the index value $\frac{\text{purity} \times \text{germination}}{100}$. This means the number of pounds per hundred of viable, pure seed. Use of the index value is of some value in computing seeding rates of small grains, grasses, clovers, soybeans and many vegetable crop seeds. The use of this method requires that standards of quality for recommended seeding rates be established. Such a basis makes it possible to adjust the seeding rate of any seed lot to the extent that it differs from the standard quality. For example, seed eligible for certification must meet certain minimum standards. If such standards are accepted in each state or region as a basis for recommended seeding rates, then each lot of seed can be measured by those standards and a seeding rate computed. A list of crop seeds showing Iowa

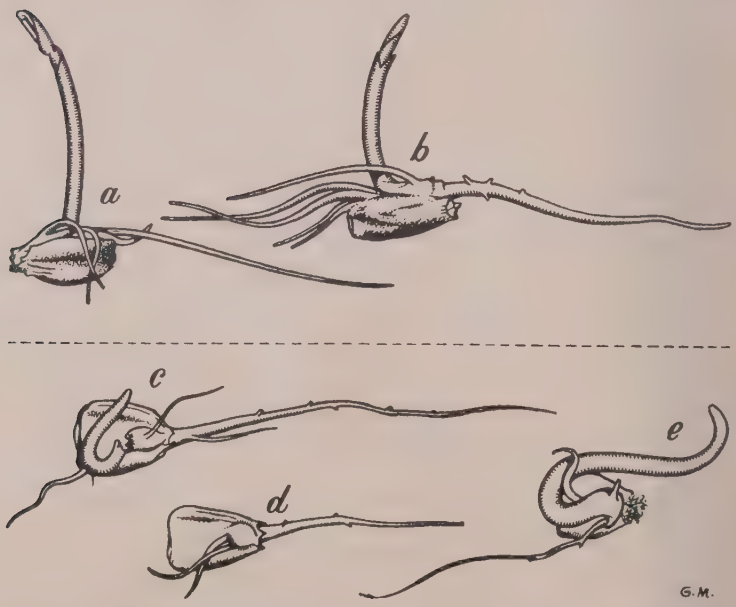

Fig. 31. Corn seedlings (natural size). a—classed as normal but without primary root; b—normal; c, d and e—abnormal.

Fig. 32. Soybean seedlings (1½ times natural size). a and b—normal; c, d and e—weak (abnormal); f, g and h—baldheads (abnormal).

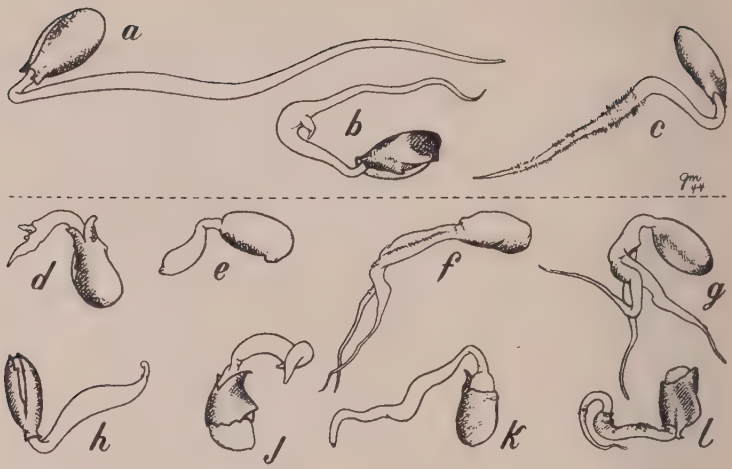

Fig. 33. Flax seedlings (4 times natural size). a, b and c—normal; d to l inclusive—types of abnormal seedlings.

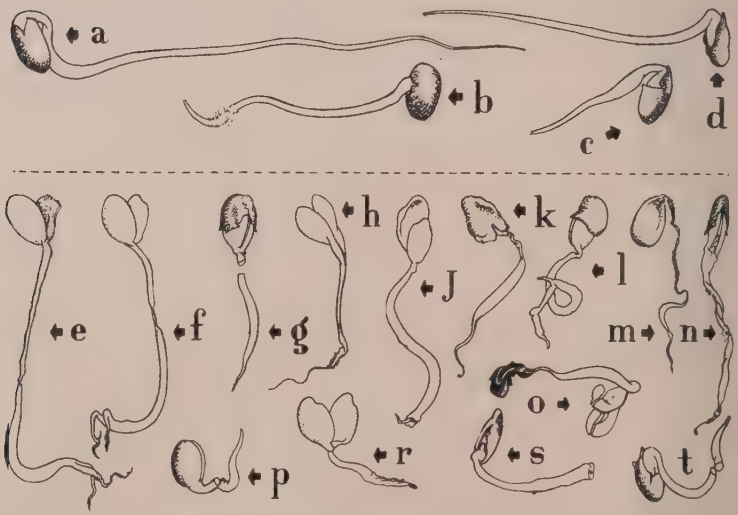

Fig. 34. Korean lespedeza seedlings (5 times natural size). a, b, c and d—normal; e to t inclusive—types of abnormal seedlings.

Fig. 35. Hemp seedlings (3 times natural size). a, b and c—normal; d to j inclusive—types of abnormal seedlings.

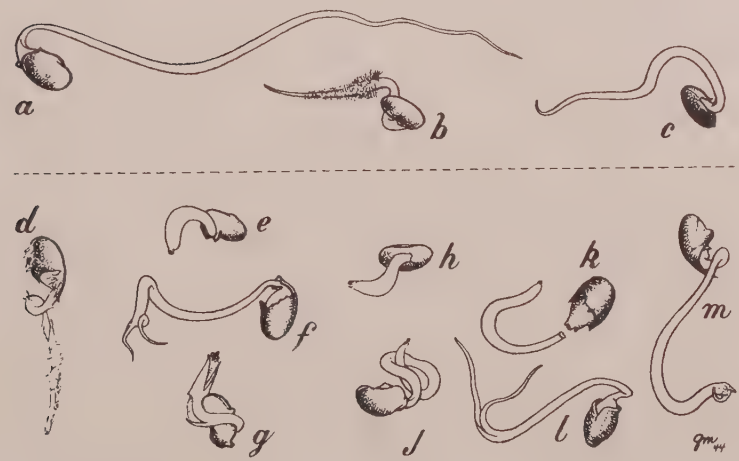

Fig. 36. Red clover seedlings (5 times natural size). a, b and c—normal; d to m inclusive—weak, malformed and broken, seedlings classed as abnormal.

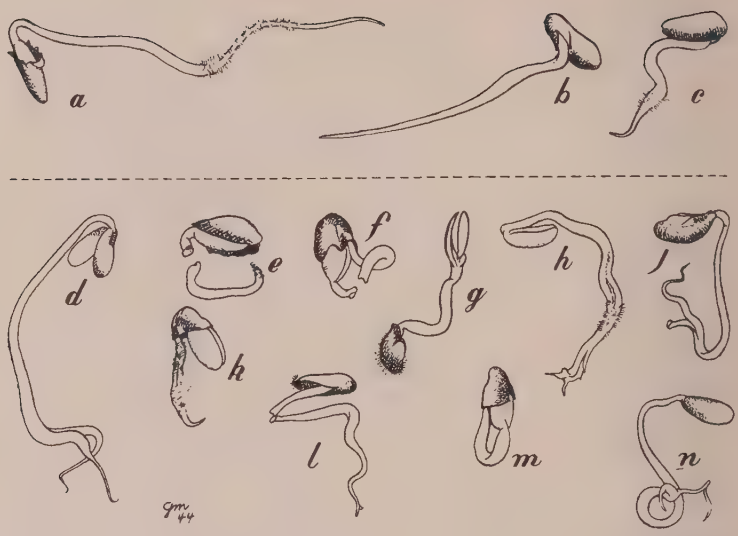

Fig. 37. Sweet clover seedlings (5 times natural size). a, b and c—normal seedlings; d to n inclusive—broken, malformed weak, seedlings classed as abnormal.

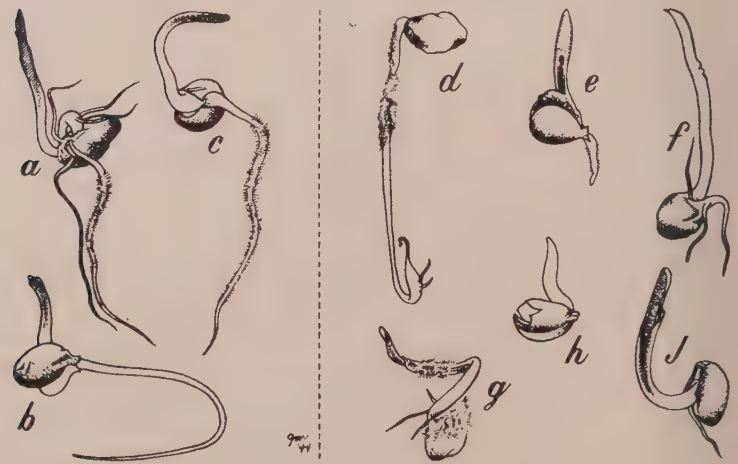

Fig. 38. Sorghum seedlings (3 times natural size). a, b and c—normal; d to j—abnormal; d—weak root and no stem; e—weak root and stem; f—stem sheath (coleoptile) but no plumule; j—plumule but practically no root.

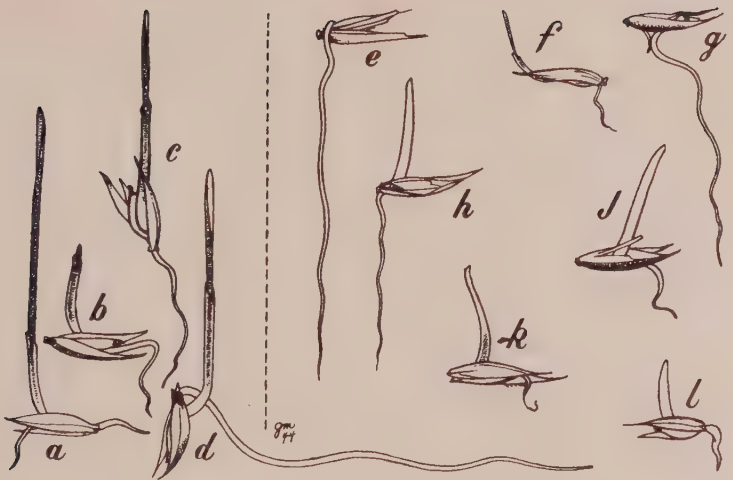

Fig. 39. Orchard grass seedlings (4 times natural size). a, b, c and d—normal; e to l inclusive—abnormal; e and g—no stem; h, j and l—no plumule, only the coleoptile.

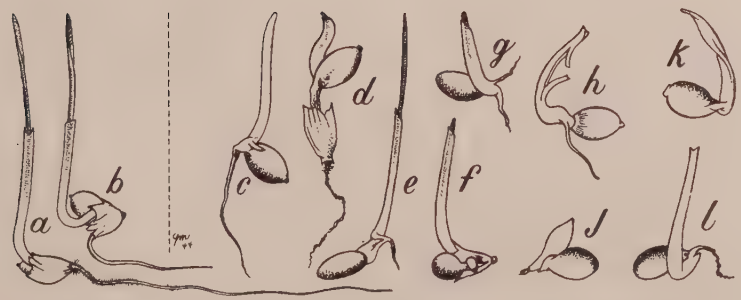

Fig. 40. Timothy seedlings (8 times natural size). a and b—normal; c to l inclusive—abnormal; e and l—no plumule, only the coleoptile.

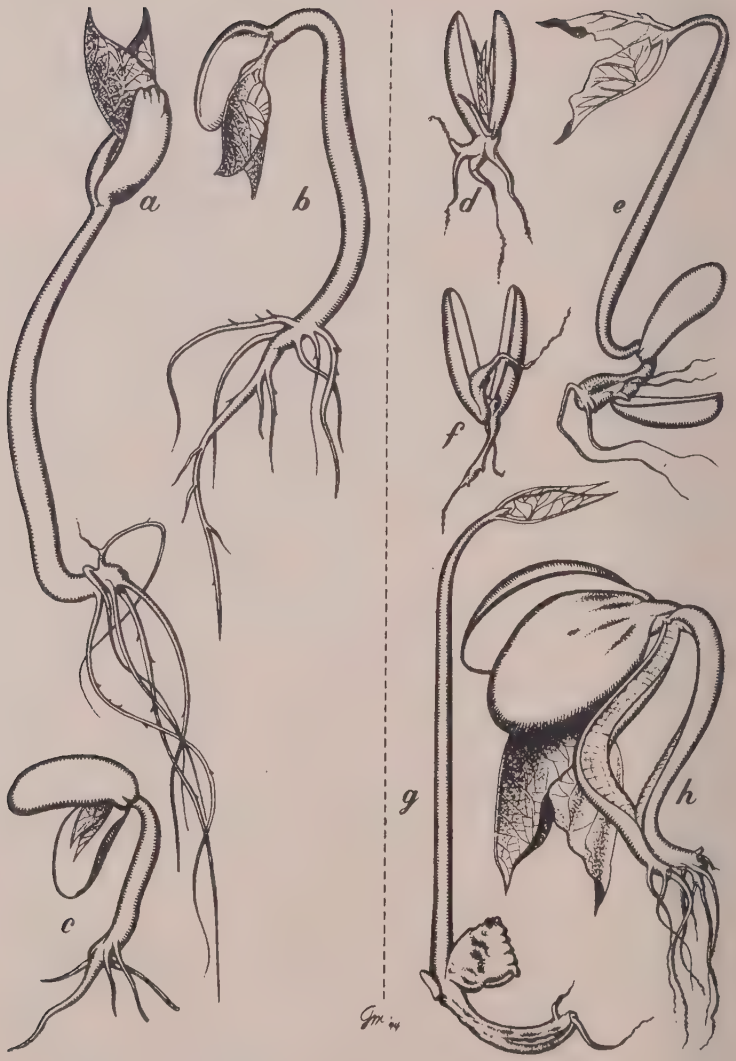

Fig. 41. Garden and lima bean seedlings (1½ times natural size). a, b and c—normal; d to h—malformed and weak (abnormal).

Fig. 42. Garden bean seedlings (1½ times natural size). j, k, l and m—types of baldheads generally classed as abnormal, but seedlings similar to l and m usually produce plants and bear fruit: n to r—malformed, abnormal seedlings.

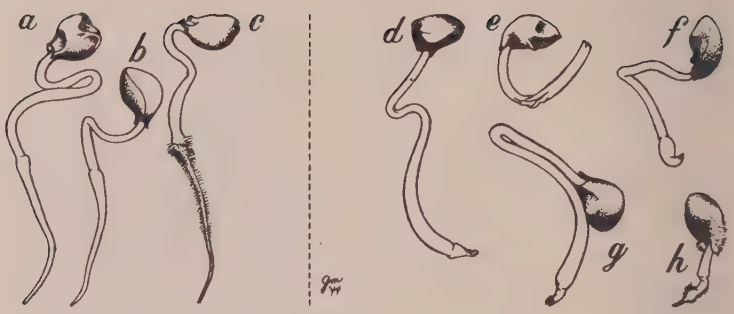

Fig. 43. Onion seedlings (5 times natural size). a, b and c—normal; d to h inclusive—abnormal.

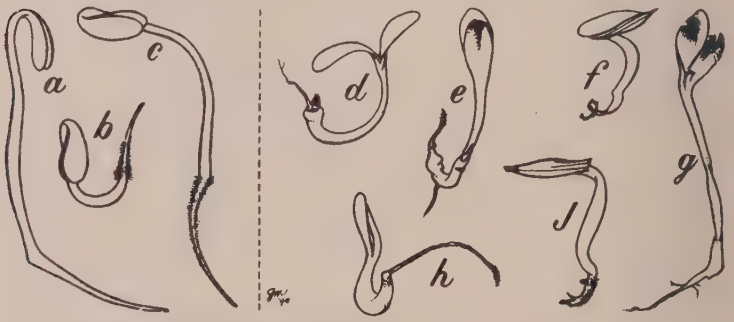

Fig. 44. Lettuce seedlings (3 times natural size). a, b and c—normal, d to j inclusive—weak, malformed, abnormal seedlings.

standards for certification and rates of seeding when sown separately follows:

Kind of crop	Minimum requirements for certification (blue tag)		Index value	Recommended seeding rate—Pounds per acre
	Purity	Germination		
Bromegrass	85	80	68.0	10*
Barley	97	90	87.3	96
Corn	100	90	90.0	8
Clover, red	97	90	87.3	8
Clover, sweet	97	90	87.3	10
Flax	97	90	87.3	45
Lespedeza, Korean	97	90	87.3	25
Oats	97	90	87.3	96
Soybeans	97	90	87.3	60
Wheat	97	90	87.3	90

* Bromegrass is not usually seeded alone in Iowa, but 10 lbs. is an average rate of seeding.

To use index value in determining rate of seeding we may take red clover as an illustration. Assume a sample of red clover with a purity of 95 percent, a germination of 70 percent and a hard seed content of 18 percent. The adjusted rate of seeding is determined by the formula,

$$\frac{\text{Standard Index Value} \times \text{Recommended Seed rate}}{\text{Index Value of Sample}} = \text{Adjusted Seeding Rate}$$

Substituting the red clover data in this formula we have

$$95 \times \frac{\frac{87.3 \times 8}{100}}{(70 + 18)}^{*} = \frac{698.4}{83.6} = 8.35 \text{ pounds}$$

For some crops such as barley, oats, flax and soybeans the number of seeds per pound varies considerably with the variety, hence a further factor affecting seeding rates is introduced. Common varieties of flax 25 years ago contained about 300 seeds per gram (136,000 per pound). In new varieties the number is sometimes less than two-thirds the earlier estimate (33). It is not advisable to make arbitary adjustments of seeding rates on the basis of number of seed per pound because of varietal differences as to stooling or branching. Other factors being the same, however, the above formula would need to be modified by inclusion of a factor that considers number of seeds per pound. The proper procedure in such a case would be to multiply the rate of seeding obtained from the above formula by the factor resulting from standard number of seeds per gram, ounce or pound (table 3) divided by the actual number per gram, ounce or pound. A specific illustration of this principle may be given with flax. Assume a sample with purity 90 percent, germination 80 percent and 200 seeds per gram of pure seed. The adjusted rate of seeding becomes

$$\frac{\frac{87.3 \times 45}{90 \times 80}}{100} \times \frac{300}{200} = \frac{87.3 \times 45 \times 100 \times 300}{90 \times 80 \times 200} =$$

$$\frac{87.3 \times 15}{16} = 81.8 \text{ lbs. per acre}$$

Further factors affecting the interpretation of seed laboratory tests in terms of practical value are (a) incidence of seed-borne organisms, (b) resistance or susceptibility to soil-borne organisms and (c) possible value of a specific seed disinfectant or protectant. Recommendations as to adjusted rates of seeding when such pathological factors are involved must necessarily be less specific. If organisms almost completely controllable by seed disinfection are carried by a seed stock then seed disinfection

*Certification standards for clovers in Iowa permit inclusion of all hard seeds as part of the germination percentage.

eliminates any consideration of that factor in planting for maximum yield. If a specific test has shown that a lot of seed is susceptible to soil-borne fungi but that a seed protectant will provide reasonable assurance of little or no injury then treatment of the seed will make further consideration of that factor practically unnecessary. On the other hand, if a given lot of corn, for example, is highly susceptible to attack by soil fungi but otherwise satisfactory and a seed protectant may be expected to insure considerable improvement in germination but not enough for maximum requirements, then planting of that lot should be delayed until a time when more favorable soil temperatures for seed germination prevail. Farmers must also consider such factors as soil moisture at the time of planting, physical condition of the soil, seedbed preparation, drainage, methods of seeding, soil reaction and general soil tilth. Such a combination of conditions makes it almost impossible to predict accurately the results that may be obtained when a given lot of seed is planted, but careful consideration of all the known factors should be superior to no consideration of any of them.

DETERMINATION OF MOISTURE IN SEEDS

The percentage of moisture in seeds has an important bearing on the keeping quality, hence it is often desirable to know the percentage of moisture before seeds are stored. Furthermore, the readiness with which seeds germinate is often determined by their moisture content. A seed laboratory should be equipped to make moisture determinations whenever necessary. This may be accomplished by the use of machines which employ the electric method that is adapted to such large seeds as soybeans and corn. A second method consists of boiling a given weight of seed in oil, collecting the vapor, condensing it and measuring the moisture removed in milliliters which gives the percentage of water in the seed on the basis of the initial sample. A third method and probably the most reliable for all kinds of seed is that of drying for 24 hours in an oven with a temperature of 100°C. (212°F.). The initial weight minus the dry weight gives the weight of water removed which divided by the initial weight gives the percentage moisture of the initial sample.

VARIATION IN SEED SAMPLES

Variation in seed samples is generally recognized as an important factor in the evaluation of seed analyses and tests, but the extent to which variations may be expected and statistical methods for the measurement of variations are not well understood.

If a sample of seed is free from all foreign matter and other seed impurities, it should be practically 100 percent pure. This seldom occurs except in the case of such seeds as corn, beans, peas, squash, pumpkin and watermelon. If repeated sub-samples are drawn from seed of such quality each should show practically 100 percent pure seed. Similarly, if a lot has 100 percent germination, each sub-sample should test 100 percent.

For the most part seed lots fall below 100 percent in both purity and germination, and the nearer the decline approaches the 50 percent point the greater the differences to be expected between a given set of sub-samples drawn from the bulk. For example, if two lots of seed have a mean purity percentage of 95 and 80, respectively, and 10 sub-samples are drawn from each lot, the difference between the replicates from the first lot will usually be less than from the second even though both lots were equally well mixed.

One of the most helpful methods of determining whether the differences between replicate sub-samples of seed are normal and to be expected is the Chi-square test for homogeneity (47). To apply this test to purity analyses and germination tests it is necessary to recognize that each sample is made up of two parts (1) the pure seed (or the germinating seed) and (2) the impurities (or the non-germinating seed). If one is known the other can be obtained by subtraction from 100. As an illustration of application let us assume that 4 separate analyses were made of a lot of alfalfa seed using approximately 5 grams of seed for each analysis. The results obtained were 95.6, 96.8, 96.0 and 97.2. The question to be answered is, "Are such results to be expected from a well-mixed lot of alfalfa seed when each sub-sample is analyzed by a uniform and standard technique?" Computation using the Chi-square test follows:

	x*	x—M	(x—M)²*
	95.6	—0.8	.64
	96.8	+0.4	.16
	96.0	—0.4	.16
	97.2	+0.8	.64
Mean=	96.4	Σ (x—M)=0	Σ (x—M)²=1.60

$$\text{Chi-square}—(\chi^2)=\frac{\Sigma(x-M)^2\times 100}{(100—M)\times M}\times \text{factor}$$

* It is assumed that each percentage of pure seed represents the number of pure seed particles per 100.

Σ = summation

M = mean percentage of purity

x — M = purity of each sub-sample minus mean percentage

Factor = number of hundreds of seeds in the working (sub-sample) sample. In table 3 the number of seeds in 5 grams of pure alfalfa seed is 2500, hence the factor becomes 25.

One helpful check on the accuracy of the mathematical computation is that the algebraic summation of the x — M column should be zero.

Substituting in the formula we have:

$$\chi^2 = \frac{1.60 \times 100}{96.4 \times 3.6} \times 25 = 11.5$$

Having obtained the Chi-square value it is necessary to consult a table of probabilities (table 7) for the distribution of Chi-square in which it will be found that for 3 degrees of freedom (number of observations minus 1) a Chi-square value of 11.34 gives a probability of 0.01. Since the calculated Chi-square value in the particular problem is 11.5 the probability value is slightly less than 0.01 (1 percent). It is arbitrarily assumed that when the probability value is less than .05 the differences in purity percentages are significant and should not be expected from well-mixed lots of seed. It is concluded from this analysis of the data that either the lot was not well mixed or the method of analysis was incorrect or non-uniform.

The Chi-square test can be applied to any number of analyses from a particular lot of seed and the results evaluated as illustrated in the preceding problem.

To illustrate further the application of the Chi-square test we may refer to germination data obtained by the author with four replicate tests of sorghum seed from a given lot using 100 seeds in each test. The calculations are as follows:

Sample no. (1)	Germination (2)	Difference from mean (3)	Square (4)
1.....	80	—1.5	2.25
2.....	76	—5.5	30.25
3.....	84	+2.5	6.25
4.....	86	+4.5	20.25
Total or Mean	M=81.5	Σ x(--M)=0.0	Σ (x—M)^2 =59.00

Step 1. Add the four percentages and divide by 4 to get mean percentage, 81.5.

Step 2. Write down in column 3 the difference between each percentage and the mean percentage.

Step 3. Write down in column 4 the square of each corresponding number in column 3 and find the total, 59.

Step 4. Multiply the mean percentage germinated (81.5) by the mean percentage failed (18.5 which is 100 — 81.5) which is 1507.75. Then Chi-square

$$(\chi^2) = \frac{100 \times 59}{1507.75} = 3.91$$

TABLE 7. DISTRIBUTION OF χ^2 WITH CORRESPONDING PROBABILITY VALUES AT DIFFERENT DEGREES OF FREEDOM *.

n	.99	.98	.95	.90	.80	.70	.50	.30	.20	.10	.05	.02	.01	.001
1	.00016	.00063	.0039	.016	.064	.15	.46	1.07	1.64	2.71	3.84	5.41	6.64	10.83
2	.020	.040	.10	.21	.45	.71	1.39	2.41	3.22	4.60	5.99	7.82	9.21	13.82
3	.12	.18	.35	.58	1.00	1.42	2.37	3.66	4.64	6.25	7.82	9.84	11.34	16.27
4	.30	.43	.71	1.06	1.65	2.20	3.36	4.88	5.99	7.78	9.49	11.67	13.28	18.46
5	.55	.75	1.14	1.61	2.34	3.00	4.35	6.06	7.29	9.24	11.07	13.39	15.09	20.52
6	.87	1.13	1.64	2.20	3.07	3.83	5.35	7.23	8.56	10.64	12.59	15.03	16.81	22.46
7	1.24	1.56	2.17	2.83	3.82	4.67	6.35	8.38	9.80	12.02	14.07	16.62	18.48	24.32
8	1.65	2.03	2.73	3.49	4.59	5.53	7.34	9.52	11.03	13.36	15.51	18.17	20.09	26.12
9	2.09	2.53	3.32	4.17	5.38	6.39	8.34	10.66	12.24	14.68	16.92	19.68	21.67	27.88
10	2.56	3.06	3.94	4.86	6.18	7.27	9.34	11.78	13.44	15.99	18.31	21.16	23.21	29.59
11	3.05	3.61	4.58	5.58	6.99	8.15	10.34	12.90	14.63	17.28	19.68	22.62	24.72	31.26
12	3.57	4.18	5.23	6.30	7.81	9.03	11.34	14.01	15.81	18.55	21.03	24.05	26.22	32.91
13	4.11	4.76	5.89	7.04	8.63	9.93	12.34	15.12	16.98	19.81	22.36	25.47	27.69	34.53
14	4.66	5.37	6.57	7.79	9.47	10.82	13.34	16.22	18.15	21.06	23.68	26.87	29.14	36.12
15	5.23	5.98	7.26	8.55	10.31	11.72	14.34	17.32	19.31	22.31	25.00	28.26	30.58	37.70
16	5.81	6.61	7.96	9.31	11.15	12.62	15.34	18.42	20.46	23.54	26.30	29.63	32.00	39.25
17	6.41	7.26	8.67	10.08	12.00	13.53	16.34	19.51	21.62	24.77	27.59	31.00	33.41	40.79
18	7.02	7.91	9.39	10.86	12.86	14.44	17.34	20.60	22.76	25.99	28.87	32.35	34.80	42.31
19	7.63	8.57	10.12	11.65	13.72	15.35	18.34	21.69	23.90	27.20	30.14	33.69	36.19	43.82
20	8.26	9.24	10.85	12.44	14.58	16.27	19.34	22.78	25.04	28.41	31.41	35.02	37.57	45.32
21	8.90	9.92	11.59	13.24	15.44	17.18	20.34	23.86	26.17	29.62	32.67	36.34	38.93	46.80
22	9.54	10.60	12.34	14.04	16.31	18.10	21.34	24.94	27.30	30.81	33.92	37.66	40.29	48.27
23	10.20	11.29	13.09	14.85	17.19	19.02	22.34	26.02	28.43	32.01	35.17	38.97	41.64	49.73
24	10.86	11.99	13.85	15.66	18.06	19.94	23.34	27.10	29.55	33.20	36.42	40.27	42.98	51.18
25	11.52	12.70	14.61	16.47	18.94	20.87	24.34	28.17	30.68	34.38	37.65	41.57	44.31	52.62
26	12.20	13.41	15.38	17.29	19.82	21.79	25.34	29.25	31.80	35.56	38.88	42.86	45.64	54.05
27	12.88	14.12	16.15	18.11	20.70	22.72	26.34	30.32	32.91	36.74	40.11	44.14	46.96	55.48
28	13.56	14.85	16.93	18.94	21.59	23.65	27.34	31.39	34.03	37.92	41.34	45.42	48.28	56.89
29	14.26	15.57	17.71	19.77	22.48	24.58	28.34	32.46	35.14	39.09	42.56	46.69	49.59	58.30
30	14.95	16.31	18.49	20.60	23.36	25.51	29.34	33.53	36.25	40.26	43.77	47.96	50.89	59.70

For larger values of n, the expression $\sqrt{2\chi^2}-\sqrt{2n-1}$ may be used as a normal deviate with unit variance.

* Reproduced by permission of R. A. Fisher.

Since there were four samples, χ^2 has three degrees of freedom and in a table of probability values a Chi-square of 3.91 would give a probability of slightly less than .30 for three degrees of freedom. It is concluded, therefore, that such differences are normal and to be expected. It is well to point out, however, that if the percentages of germination in column two had been based on 400 instead of 100 seeds, the Chi-square would be four times as great: namely, 15.61 which would give a probability of almost .001. In other words, the differences would be highly significant and not expected from well-mixed samples of seed.

The appropriate formula for Chi-square when analyzing the data from germination tests is $\frac{S\Sigma (P - \bar{P})^2}{\overline{PQ}}$ where S = number of seeds tested in each sample, $\Sigma (P - \bar{P})^2$ = sum of squares of the differences between each percentage germinated ($\bar{P}$) and the mean percentage germinated ($\bar{P}$). Q. = 100 − P = mean percentage failed.

TOLERANCE AS APPLIED TO SEED TESTING

The Federal Seed Act (46) and most state seed acts recognize that consideration must be given to differences in samples drawn at different times and by different people from a given bulk lot of seed or from different portions of a lot. For example, a shipper or vendor may draw what he considers a representative sample from a bulk lot, have it tested and the lot labeled accordingly. A purchaser who receives part or all of the lot may then draw a sample for a test or a seed inspector may draw a sample. Comparison of the data on the label is then made with the data obtained later. A basis of evaluating the two sets of data is necessary either for payment by the purchaser or for decision as to correct or incorrect labeling of the seed.

To facilitate decisions, rules of tolerance have been established for purity, weed and crop seed content, inert matter and germination. These tolerances are more liberal than would be provided by the Chi-square test for homogeneity, using even a 1 percent probability as the point at which differences are considered significant. The formula for determining tolerance in purity percentages is $T = 0.6 + 0.2 \times \frac{(a \times b)}{100)}$ where T = tolerance, a = percentage pure seed and b = 100 − a. To illustrate the use of the formula assume a lot of seed is labeled 95.6 percent pure seed and the inspector's sample shows 94 percent pure. The computation is $T = 0.6 + \frac{(0.2 \times 94 \times 6)}{100} = 1.728$;

94 + T = 94 + 1.728 = 95.728 which is greater than 95.6 as claimed, hence the lot would not be considered mislabeled.

This formula is used for all kinds of seed except such chaffy grasses as bluegrass, orchard grass, bromegrass, wheat grasses and redtop. For chaffy grasses an additional tolerance is allowed; namely, by adding to the regular tolerance described, the product obtained by multiplying "T" by the lesser of "a" and "b" divided by 100. To illustrate the application assume a lot of bluegrass labeled 90 percent pure seed and the inspector's sample shows 85. First the regular tolerance is computed:

$$T = 0.6 + \left(0.2 \times \frac{85 \times 15}{100}\right) = 3.15.$$

The additional tolerance is $T \times \frac{15}{100} = .47$. The total tolerance is then 3.15 + .47 = 3.62 and 85 + 3.62 = 88.62 which is less than 90, the percent claimed, and the lot would be considered as mislabeled.

Tolerance for weed seeds, other crop seeds and inert matter is computed by the formula $T = 0.2 + \left(0.2 \times \frac{a \times b}{100}\right)$.

Recognized tolerances applicable to percentage of germination and to the sum of the percentages of germination and of hard seed are as follows:

Found by Test	Tolerance
96 or over	5
90 or over but less than 96	6
80 or over but less than 90	7
70 or over but less than 80	8
60 or over but less than 70	9
Less than 60	10

The application of these tolerances is as follows: Assume a lot labeled 90 percent germination and the inspector's sample gives 83 percent. The tolerance is applied to what is found (83) which is 7, and 83 + 7 = 90 hence the lot would not be considered mislabeled.

It is also necessary to have tolerances for evaluating the number of noxious weed seeds in a given lot as shown on a label. For example, if a lot is claimed to have two noxious weed seeds per ounce of seed and a subsequent analysis shows more than two per ounce, how many more than two would be allowable? According to the recognized tolerance a total of six in an examination of a 1 ounce sample would not justify a claim of mislabeling as shown at the top of the next page.

TOLERANCE FOR NOXIOUS WEED SEEDS IN ANY GIVEN WEIGHT

Claimed	Found	Claimed	Found
0	2	11	18
1	4	12	20
2	6	13	21
3	8	14	22
4	9	15	23
5	11	16	24
6	12	17	25
7	13	18	27
8	14	19	28
9	16	20	29
10	17	21	30

The numbers shown in columns two and four indicate the quantity above which a lot is considered as mislabeled if the respective quantities in columns one and three are claimed on the label. Vendors should not, however, use these tolerances by putting on a label a number of weed seeds correspondingly less than actually found by their tests. Such a practice inevitably ends in difficulty with law enforcing agencies.

SEED LAWS

Iowa and other states have found it necessary to enact laws regulating the sale of seeds within their respective boundaries. These laws known as "acts" are enforced by the secretary or commissioner of agriculture. In addition, congress has enacted a Federal Seed Act enforced by the United States Department of Agriculture. The act regulates the movement of seed in interstate commerce and controls the shipment of seed from other countries into any part of the United States and its outlying possessions. These acts are essentially labeling laws in that they require all seed lots to be labeled so as to show the purity, germination and weed seed content and in some cases the origin.

DIGEST OF THE IOWA SEED LAW [3]

The new Iowa Seed Law has four main features which need discussion, namely, (a) list of noxious weeds, (b) labeling requirements, (c) prohibited sales and (d) exemptions.

Noxious weed seeds are divided into two classes as follows:

Primary

Common Name	Botanical Name
Quack grass	*Agropyron repens*
Canada thistle	*Cirsium arvense*
Perennial sow thistle	*Sonchus arvensis*
Perennial peppergrass	*Lepidium draba*
Field bindweed (creeping Jennie)	*Convolvulus arvensis*
Horse nettle	*Solanum carolinense*
Leafy spurge	*Euphorbia esula*
Russian knapweed	*Centaurea repens*

[3]Enforcement of the Iowa Seed Law is under the jurisdiction of the State Secretary of Agriculture at Des Moines. Inquiries concerning this law should be directed to the State Department of Agriculture, Des Moines, Iowa.

Secondary

Common Name	Botanical Name
Wild carrot	*Daucus carota*
Sour dock	*Rumex crispus*
Smooth dock	*Rumex altissimus*
Sheep sorrel	*Rumex acetosella*
Butterprint	*Abutilon theophrasti*
Mustards	*Brassica spp.*
Cocklebur	*Xanthium commune*
Buckhorn	*Plantago lanceolata*
Dodders	*Cuscuta spp.*

All seed dealers and farmers who sell off their own farms are required to supply the following information on a label, if in bags, or placard if sold in bulk:

(1) Commonly accepted name of (a) kind, or (b) kind and variety or (c) kind and type of each agricultural seed component in excess of 5 percent of the whole and the percentage by weight of each in the order of its preponderance.

(2) Lot number or other lot identification.

(3) Origin, if known, of alfalfa and red clover. If the origin is unknown, that fact shall be stated.

(4) Percentage by weight of all weed seeds.

(5) The name and approximate number of each kind of secondary noxious weed seed, per ounce in groups (a), (b) and (c), and per pound in group (d), when present singly or collectively in excess of—

(a) Five seeds or bulblets per ounce of redtop, bluegrass, Bermuda grass, timothy, orchard grass, fescues (except meadow fescue), alsike and white clover, reed canary grass and other agricultural seeds of similar size and weight, or mixture within this group;

(b) Three seeds or bulblets per ounce of ryegrass, meadow fescue, foxtail millet, alfalfa, red clover, sweet clover, lespedeza, smooth brome, crimson clover, *Brassica* spp., flax, *Agropyron* spp. and other agricultural seeds of similar size and weight, or mixtures within this group, or of this group with (a);

(c) One seed or bulblet per ounce of proso, sudan grass and other agricultural seeds of similar size and weight, or mixtures not specified in (a), (b) or (d);

(d) Five seeds or bulblets per pound of wheat, oats, rye, barley, buckwheat, sorghum (except sudan grass), vetches, soybeans and other agricultural seeds of a size and weight similar to or greater than those with-

in this group. All determinations of noxious weed seeds are subject to tolerances and methods of determination prescribed in the rules and regulations under this act.

(6) Percentage by weight of agricultural seeds other than those required to be named on the label.

(7) Percentage by weight of inert matter.

(8) For each named agricultural seed (a) percentage of germination exclusive of hard seed, (b) percentage of hard seed, if present and (c) the calendar month and year the test was completed to determine such percentages. Following (a) and (b) the additional statement "total germination and hard seed" may be stated as such, if desired.

(9) Warning as to danger from poisoning in the case of treated seed if compound is used which is poisonous to man or farm animals.

(10) Name and address of the person who labeled said seed, or who sells, offers or exposes said seed for sale within this state.

SALE BY GROWER ON HIS OWN FARM

A grower who sells seed on his own farm either in bulk or in containers may be exempt from the labeling provisions referred to provided that either a placard is displayed or a written or printed statement is supplied to the purchaser with the following information:

(1) The percentage germination and of hard seeds of the seed being sold, together with the calendar month and year said seed was tested to determine the percentages.

(2) The kind and number per ounce or pound of all secondary noxious weed seeds in the lot when in excess of the amounts given under b (5) page 579.

(3) A guarantee that no primary noxious weed seeds are present in accordance with recognized tolerances.

PROHIBITED SALES

It is unlawful for any person to sell, offer for sale or expose for sale within Iowa:

(1) Any agricultural seed:

(a) Unless the test to determine the percentage of germination required by Section 3 shall have been completed within a 9-month period, exclusive of the calendar month in which the test was completed, immediately prior to sale, exposure for sale, or offering for sale or transportation.

(b) Not labeled in accordance with the provisions of the act, or having a false or misleading labeling.

(c) Pertaining to which there has been a false or misleading advertisement.

(d) Containing any primary noxious weed seeds subject to tolerances and methods of determination prescribed in the rules and regulations.

(e) Containing more than 3 percent weed seeds.

(2) Screenings of any agricultural seed unless it is stated on the label, if in containers, or on the invoice, if in bulk, that they are not intended for seeding purposes.

It is further unlawful for any person—

(1) To detach, alter, deface or destroy any label provided for in the act.

(2) To disseminate any false or misleading advertisement concerning agricultural seed in any manner or by any means.

(3) To hinder or obstruct in any way any authorized person in the performance of his duties.

(4) To fail to comply with a "stop sale" order.

EXEMPTIONS

Labeling provisions and prohibitions do not apply to—

(1) Seed or grain not intended for sowing purposes.

(2) Seed in storage in or consigned to a seed cleaning establishment for cleaning or processing.

Sale of hybrid seed corn and seed potatoes is regulated as follows:

Section 5—It shall be unlawful for any person to sell, offer or expose for sale or falsely mark or tag, within the state of Iowa any seed corn as hybrid unless it represents the first generation of a cross between strains of different parentage and involving inbred lines of corn and (or) their combinations. Any corn sold as "hybrid" shall have plainly printed or marked on the label or container in which such corn is sold the identifying symbols or numbers, clearly indicating the specific combination. The cross mentioned above shall be produced by cross fertilization, controlled either by hand or detasseling at the proper time.

Section 7—It is hereby established that a certification system is essential to the supply of good seed potato stocks for the state of Iowa and that long usage of a blue tag attached to bags containing certified seed by authorities in states where certified seed potatoes are produced has become identified in the public mind as evidence of superior quality and of official certification.

It shall be unlawful for any person to sell, offer for sale or expose for sale in the state of Iowa—

(1) Any seed potatoes with a blue tag attached, unless same are certified.

(2) Any seed potatoes as "certified" unless—

(a) Each bag bears a label blue in color with the word "certified" thereon.

(b) Such seed has been certified by a duly constituted state authority or state association in the state in which the seed was produced; said state authority or association to be recognized by the Iowa secretary of agriculture.

THE FEDERAL SEED ACT OF 1939 [4]

The Federal Seed Act was designated to regulate both interstate shipments of seed and seed importations (29).

Seed moving in interstate commerce must be labeled to show (a) kind or kind and variety or kind and type for each agricultural seed component present in excess of 5 percent of the whole and the percentage by weight of each, (b) lot number or other identification, (c) origin of red clover and alfalfa or of any other seed as may be required by the Secretary of Agriculture, (d) percentage by weight of all weed seeds, including noxious weed seeds, (e) kinds and rate of occurrence of noxious weed seeds in accordance with requirements of the destination state, (f) percentage by weight of other agricultural seed, (g) percentage by weight of inert matter, (h) percentage of germination and of hard seed separately of each kind, or kind and variety or kind and type in excess of 5 percent of the whole, together with the month and year tested and (i) the name and address of the person who ships, transports or delivers for transportation said seed in interstate commerce or the person to whom the seed is sold or shipped for resale together with a code designation approved by the Secretary of Agriculture. Two further important requirements under the act are that (a) screenings cannot be shipped in interstate commerce unless they are not intended for seeding purposes and are labeled or invoiced with the words "Screenings for processing—not for seeding," and (b) seed for cleaning or processing must be labeled or invoiced with the words "Seed for processing."

The more important provisions as to seed importations deal with prohibited entries and the staining of alfalfa and red clover

[4] Enforcement of the Federal Seed Act is under the jurisdiction of the United States Department of Agriculture. Rules and regulations relative to methods of testing and labeling of seeds to meet the requirements of the act have been promulgated by the Department. These rules and methods should be followed by all who label seed that is to move in interstate commerce. Inquiries relative to the act should be directed to the Seed Division, Grain Products Branch, War Food Administration, Washington, D. C.

seed to indicate the country of origin. Seed classed as unfit for seeding purposes is denied entry into the United States and includes: (a) seeds containing noxious weed seeds in excess of 1 in 10, 25, or 100 grams of seed, depending on the respective size of crop seed, (b) seed containing more than 2 percent by weight of weed seed and (c) seed containing less than 75 percent of pure live seed. Weed seeds classed as noxious in imported seed are those of Canada thistle (*Cirsium arvense*), quack grass (*Agropyon repens*), white top (*Lepidium draba, Lepidium repens* and *Hymenophysa pubescens*), Johnson grass (*Holcus halepense*), bindweed (*Convolvulus arvensis*), Russian knapweed (*Centaurea picris*), perennial sow thistle (*Sonchus arvensis*), leafy spurge (*Euphorbia esula*) and dodders (*Cuscuta spp.*).

The United States Department of Agriculture in cooperation with several state experiment stations has determined that strains of alfalfa and red clover seed of foreign origin other than from the Dominion of Canada are unadapted for general use in the United States. To protect farmers the Federal Seed Act requires that imported seed of alfalfa or red clover in each container be stained to indicate origin as follows:

(a) Seed grown in any foreign country other than the countries of South America and the Dominion of Canada must have 10 percent stained red;

(b) Seed grown in any of the countries of South America must have 10 percent stained orange-red;

(c) Seed grown in the Dominion of Canada must have 1 percent stained violet;

(d) Seed must have 10 percent stained red if:

(1) the origin is unestablished;

(2) the origin is such as to require different colors; and

(3) seed of foreign origin is commingled with seed of the same kind grown in the United States.

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